```
=> b hcaplus
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FILE COVERS 1907 - 9 Jul 2004  VOL 141 ISS 3
FILE LAST UPDATED: 8 Jul 2004  (20040708/ED)

 This file contains CAS Registry Numbers for easy and accurate
 substance identification.

'OBI' IS DEFAULT SEARCH FIELD FOR 'HCAPLUS' FILE

=> => d que l6
L1  (   1022897)SEA FILE=HCAPLUS ABB=ON  PLU=ON  PROTEIN?/CT,CW
L2  (    192277)SEA FILE=HCAPLUS ABB=ON  PLU=ON  L1(L)(PREP+NT OR PROC+NT)/RL
L3  (     11206)SEA FILE=HCAPLUS ABB=ON  PLU=ON  "PROTEIN FOLDING"/CT
L4  (      2966)SEA FILE=HCAPLUS ABB=ON  PLU=ON  L2 AND L3
L5  (       151)SEA FILE=HCAPLUS ABB=ON  PLU=ON  L4 AND P/DT
L6            18 SEA FILE=HCAPLUS ABB=ON  PLU=ON  L5 AND (PY<=1995 OR PRY<=1995
                 OR AY<=1995)


=> b biosis
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FILE COVERS 1969 TO DATE.
CAS REGISTRY NUMBERS AND CHEMICAL NAMES (CNs) PRESENT
FROM JANUARY 1969 TO DATE.

RECORDS LAST ADDED: 7 July 2004 (20040707/ED)

FILE RELOADED: 19 October 2003.

=> => d que l12
L7  (    400937)SEA FILE=BIOSIS ABB=ON  PLU=ON  PROTEIN?/CT,CW
L8  (     36972)SEA FILE=BIOSIS ABB=ON  PLU=ON  PURIFICATION(W)METHOD
L9  (      5099)SEA FILE=BIOSIS ABB=ON  PLU=ON  L8 AND L7
L10 (       205)SEA FILE=BIOSIS ABB=ON  PLU=ON  L9 AND ?SOLUBIL?
L11 (        24)SEA FILE=BIOSIS ABB=ON  PLU=ON  REFOLD? AND L10
L12           1 SEA FILE=BIOSIS ABB=ON  PLU=ON  L11 AND PY<=1995


=> b embase
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FILE COVERS 1974 TO 1 Jul 2004 (20040701/ED)

EMBASE has been reloaded. Enter HELP RLOAD for details.

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=> => d que l20
L13 (     24210)SEA FILE=EMBASE ABB=ON  PLU=ON  "PROTEIN PURIFICATION"/CT
L14 (    246440)SEA FILE=EMBASE ABB=ON  PLU=ON  (RECOMB? OR ENGINEER?)
L15 (      5086)SEA FILE=EMBASE ABB=ON  PLU=ON  L14 AND L13
L16 (     20401)SEA FILE=EMBASE ABB=ON  PLU=ON  "PROTEIN FOLDING"/CT
L17 (       305)SEA FILE=EMBASE ABB=ON  PLU=ON  L15 AND L16
L18 (        58)SEA FILE=EMBASE ABB=ON  PLU=ON  L17 AND PY<=1995
L19 (    159116)SEA FILE=EMBASE ABB=ON  PLU=ON  ?SOLUBIL? OR ?FORMULA?
L20           13 SEA FILE=EMBASE ABB=ON  PLU=ON  L19 AND L18


=> b wpix
FILE 'WPIX' ENTERED AT 09:46:49 ON 09 JUL 2004
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FILE LAST UPDATED:          2 JUL 2004      <20040702/UP>
MOST RECENT DERWENT UPDATE:     200442        <200442/DW>
DERWENT WORLD PATENTS INDEX SUBSCRIBER FILE, COVERS 1963 TO DATE

>>> FOR A COPY OF THE DERWENT WORLD PATENTS INDEX STN USER GUIDE,
    PLEASE VISIT:
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    http://thomsonderwent.com/coverage/latestupdates/           <<<

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    http://www.thomsonscientific.com/litalert           <<<

>>> THE DISPLAY LAYOUT HAS BEEN CHANGED TO ACCOMODATE THE
    NEW FORMAT GERMAN PATENT APPLICATION AND PUBLICATION
    NUMBERS. SEE ALSO:
    http://www.stn-international.de/archive/stnews/news0104.pdf <<<

=> => d que l27
L21 (      4315)SEA FILE=WPIX ABB=ON  PLU=ON  (B04-B04A5 OR B04-N03)/MC
L22 (     12468)SEA FILE=WPIX ABB=ON  PLU=ON  D05-H13/MC
L23 (       267)SEA FILE=WPIX ABB=ON  PLU=ON  L21 AND L22
L24 (      7491)SEA FILE=WPIX ABB=ON  PLU=ON  C12P021-00/IC
L25 (        47)SEA FILE=WPIX ABB=ON  PLU=ON  L23 AND L24
L26 (    238921)SEA FILE=WPIX ABB=ON  PLU=ON  ?FOLD?/BIX
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L27 3 SEA FILE=WPIX ABB=ON PLU=ON L25 AND L26

=> dup rem l12 l6 l20 l27
FILE 'BIOSIS' ENTERED AT 09:48:22 ON 09 JUL 2004
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PROCESSING COMPLETED FOR L12
PROCESSING COMPLETED FOR L6
PROCESSING COMPLETED FOR L20
PROCESSING COMPLETED FOR L27
L28 35 DUP REM L12 L6 L20 L27 (0 DUPLICATES REMOVED)

=> => b hcaplus, biosis, embase, wpix
FILE 'HCAPLUS' ENTERED AT 09:53:48 ON 09 JUL 2004
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=> d all l28 2 3 4 6 7 8 9 13 15 16 20 21 22 25 29 33 34

L28 ANSWER 2 OF 35 HCAPLUS COPYRIGHT 2004 ACS on STN
AN 2001:863534 HCAPLUS
DN 135:362531
ED Entered STN: 29 Nov 2001
TI Method of solubilizing, purifying, and refolding protein
IN Dorin, Glenn J.; Arve, Bo H.; Pattison, Gregory L.; Halenbeck, Robert F.; Johnson, Kirk; Chen, Bao-lu; Rana, Rajsharan K.; Hoba, Maninder S.; Madani, Hassan; Tsang, Michael; Gustafson, Mark E.; Bild, Gary S.; Johnson, Gary V.
PA Chiron Corporation, USA; G.D. Searle and Co.
SO U.S., 50 pp., Cont.-in-part of U.S. Ser. No. 473,668, abandoned.
CODEN: USXXAM
DT **Patent**
LA English
IC A23J001-00
NCL 530412000
CC 63-3 (Pharmaceuticals)
FAN.CNT 12

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE

PI US 6323326 B1 20011127 US 1999-973211 19990611 <--
WO 9640784 A2 19961219 WO 1996-US9980 19960607 <--
WO 9640784 A3 19970313
W: AL, AM, AT, AU, AZ, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EE, ES, FI, GB, GE, HU, IL, IS, JP, KE, KG, KP, KR, KZ, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG
RW: KE, LS, MW, SD, SZ, UG, AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM
US 5888968 A 19990330 US 1996-734997 19961022 <--
US 6319896 B1 20011120 US 1999-443098 19991118 <--
AU 759412 B2 20030417 AU 2000-20611 20000302
US 2002137884 A1 20020926 US 2001-996588 20011130 <--
JP 2004083591 A2 20040318 JP 2003-326585 20030918 <--
PRAI US 1995-473668 B2 19950607 <--
US 1995-477677 A2 19950607 <--
WO 1996-US9980 W 19960607
US 1995-473688 B1 19950607 <--
AU 1996-64770 A3 19960607
JP 1997-502126 A3 19960607
US 1999-973211 A3 19990611
US 1999-443099 B1 19991118

AB A method of modifying protein solubility employs polyionic polymers. These facilitate the solubilization, formulation, purification and refolding of proteins especially incorrectly folded proteins and aggregated proteins. Compns. are described that are suitable for formulating TFPI. The compns. allow preparation of pharmaceutically acceptable compns. of TFPI at concns. above 0.2 mg/mL and above 10 mg/mL.

ST protein solubilization purifn refolding

IT Denaturants
(chaotropic; method of solubilizing, purifying, and refolding protein)

IT Ion exchangers
Polyelectrolytes
Purification
Solubility
Solubilization
(method of solubilizing, purifying, and refolding protein)

IT Polyphosphates
RL: DEV (Device component use); PEP (Physical, engineering or chemical process); PROC (Process); USES (Uses)
(method of solubilizing, purifying, and refolding protein)

IT **Proteins, general, biological studies**
RL: **PEP (Physical, engineering or chemical process)**; **PUR (Purification or recovery)**; THU (Therapeutic use); BIOL (Biological study); **PREP (Preparation)**; **PROC (Process)**; USES (Uses)
(method of solubilizing, purifying, and refolding protein)

IT **Protein folding**
(refolding; method of solubilizing, purifying, and refolding protein)

IT Polysaccharides, uses
RL: DEV (Device component use); PEP (Physical, engineering or chemical process); PROC (Process); USES (Uses)
(sulfated; method of solubilizing, purifying, and refolding protein)

IT 9005-49-6, Heparin, uses 9042-14-2, Dextran sulfate
RL: DEV (Device component use); PEP (Physical, engineering or chemical process); PROC (Process); USES (Uses)
(method of solubilizing, purifying, and refolding protein)

IT 194554-71-7P, Tissue factor inhibitor
RL: PEP (Physical, engineering or chemical process); PUR (Purification or recovery); THU (Therapeutic use); BIOL (Biological study); PREP

(Preparation); PROC (Process); USES (Uses)
(method of solubilizing, purifying, and refolding protein)
IT 194554-71-7P, Tissue factor inhibitor
RL: PEP (Physical, engineering or chemical process); PUR (Purification or recovery); THU (Therapeutic use); BIOL (Biological study); PREP (Preparation); PROC (Process); USES (Uses)
(method of solubilizing, purifying, and refolding protein)
RE.CNT 19 THERE ARE 19 CITED REFERENCES AVAILABLE FOR THIS RECORD
RE
(1) Anon; EP 0131864 1985 HCAPLUS
(2) Anon; EP 0325691 1989 HCAPLUS
(3) Anon; EP 0473564 A1 1992 HCAPLUS
(4) Anon; EP 0559632 A2 1993 HCAPLUS
(5) Bernhard, F; Biotechnology and Bioengineering 1993, V41, P3
(6) Dabora; The Journal of Biological Chemistry 1991, V266(35), P23637 HCAPLUS
(7) Diaz-Collier; US 5212091 1993 HCAPLUS
(8) Fanning; US 5051497 1991 HCAPLUS
(9) Harenberg, J; Blood Coagulation and Fibrinolysis 1995, V6(Suppl 1)
(10) Josic; J Chromatography 1993, V632, P1 HCAPLUS
(11) Lehninger; Principles of Biochemistry 1993, P113
(12) Mark, E; Protein Expression and Purification 1994, V5, P233
(13) Petersen; US 5378614 1995 HCAPLUS
(14) Rainer, R; Protein Engineering:Principles and Practice, Chapter 10 1996, P283
(15) Rausch; US 4766224 1988 HCAPLUS
(16) Rudolph & Lilie; FASEB J V10, P49
(17) Sprecher; PNAS 1994, V91(8), P3353 HCAPLUS
(18) Tuddenham; Journal of Laboratory and Clinical Medicine 1979, V93(1), P40 HCAPLUS
(19) Wun; US 5466783 1995 HCAPLUS

L28 ANSWER 3 OF 35 HCAPLUS COPYRIGHT 2004 ACS on STN
AN 2000:909119 HCAPLUS
DN 134:70368
ED Entered STN: 28 Dec 2000
TI Recombinant production of immunoglobulin-like domains in prokaryotic cells for use in immunization
IN Ward, E. Sally; Kim, Jin-Kyoo
PA Board of Regents, the University of Texas System, USA
SO U.S., 55 pp., Cont.-in-part of U.S. Ser. No. 963,333, abandoned.
CODEN: USXXAM
DT **Patent**
LA English
IC ICM C12P021-06
NCL 435069100
CC 15-3 (Immunochemistry)
Section cross-reference(s): 3, 16
FAN.CNT 3

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	US 6165745	A	20001226	US 1994-341560	19941117 <--
PRAI	US 1992-873930	B2	19920424 <--		
	US 1992-963333	B2	19921019 <--		

AB Disclosed are recombinant vectors encoding Ig-like domains and portions thereof, such as T-cell variable domains, antibody Fc-hinge fragments, subfragments and mutant domains with reduced biol. half lives. Methods of producing large quantities of such domains, heterodimers, and fusion proteins following expression and secretion by Gram-neg. bacteria are also reported. Single chain T-cell receptors, which are folded into β-pleated sheet structures similar to those of Ig variable domains

were prepared with Escherichia coli. Antibody Fc and Fc-hinge domains were found to have the same in vivo stability as intact antibodies. Specific residues contributing to antibody stability were identified and CH2 and CH3 domains engineered to have reduced in vivo half lives were prepared

ST Ig domain Gram neg bacteria heterologous prodn; antibody Fc stability prodn prokaryote

IT Ricins
RL: ARG (Analytical reagent use); ANST (Analytical study); USES (Uses)
(A, conjugates, to antibody, in immunotoxin technol.; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT Protein motifs
(CH2-CH3, of Fc-hinge, modifying natural residues in; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT Protein motifs
(Fc-hinge domains, of antibody, impaired SpA binding to modified; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT β-Sheet
(T-cell receptors folded into; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT **Protein folding**
(T-cell receptors, β-sheet structures; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT Gene, animal
RL: BUU (Biological use, unclassified); PRP (Properties); BIOL (Biological study); USES (Uses)
(VαTCR, for T-cell receptor variable domain; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT Gene, animal
RL: BUU (Biological use, unclassified); PRP (Properties); BIOL (Biological study); USES (Uses)
(VβTCR, for T-cell receptor variable domain; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT Escherichia coli
Gram-negative bacteria
Prokaryote
Serratia marcescens
(as expression host; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT Toxins
RL: ARG (Analytical reagent use); ANST (Analytical study); USES (Uses)
(conjugated to antibody; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT Genetic vectors
(encoding Ig-like domains; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT Imaging
(fluorescent, or with paramagnetic ion, or with radioactive agent, of antibody; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT **Proteins, specific or class**
RL: **BPR (Biological process)**; BSU (Biological study, unclassified); BIOL (Biological study); **PROC (Process)**
(gene spaA, impaired binding of, to mutated antibody Fc-hinge domain; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT Fermentation
(protein; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT Genetic engineering

Immunization
(recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT Immunoglobulins
RL: BPN (Biosynthetic preparation); BUU (Biological use, unclassified); BIOL (Biological study); PREP (Preparation); USES (Uses)
(recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT TCR (T cell receptors)
RL: BUU (Biological use, unclassified); PRP (Properties); THU (Therapeutic use); BIOL (Biological study); USES (Uses)
(recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT Plasmid vectors
(scVαVβpelBHis; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT Mutagenesis
(site-directed, Fc-hinge domain mutation introduced by; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT Mutation
(substitution, in CH2-CH3, of Fc-hinge domain; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT Antibodies
RL: BPN (Biosynthetic preparation); BUU (Biological use, unclassified); BIOL (Biological study); PREP (Preparation); USES (Uses)
(with reduced half lives, conjugated to imaging agent; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT 314783-03-4, TCR (T cell receptor) (human)
RL: BUU (Biological use, unclassified); PRP (Properties); BIOL (Biological study); USES (Uses)
(amino acid sequence; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT 56-41-7, Alanine, biological studies
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(histidine at 310 and 433 substituted with; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT 152989-51-0P 152989-53-2P 152989-55-4P 152989-57-6P
RL: BPN (Biosynthetic preparation); PRP (Properties); THU (Therapeutic use); BIOL (Biological study); PREP (Preparation); USES (Uses)
(recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT 152989-50-9 152989-52-1 152989-54-3 152989-56-5, DNA (synthetic plasmid scVαVβpelBHis)
RL: BUU (Biological use, unclassified); PRP (Properties); THU (Therapeutic use); BIOL (Biological study); USES (Uses)
(recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT 73-32-5, Isoleucine, biological studies
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(substitution of, in position 253; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT 56-85-9, Glutamine, biological studies
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(substitution of, in position 311, with asparagine; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT 70-47-3, Asparagine, biological studies
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(substitution of, in position 434, with glutamine; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT 71-00-1, Histidine, biological studies

RL: BSU (Biological study, unclassified); BIOL (Biological study)
(substitution of, in positions 310 and 433, with alanine; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT 314783-27-2, 1: PN: US6165745 SEQID: 9 unclaimed DNA 314783-28-3, 2: PN: US6165745 SEQID: 10 unclaimed DNA 314783-29-4, 3: PN: US6165745 SEQID: 11 unclaimed DNA 314783-30-7, 4: PN: US6165745 SEQID: 12 unclaimed DNA 314783-31-8, 5: PN: US6165745 SEQID: 13 unclaimed DNA 314783-32-9, 6: PN: US6165745 SEQID: 14 unclaimed DNA 314783-33-0, 7: PN: US6165745 SEQID: 15 unclaimed DNA 314783-34-1, 8: PN: US6165745 SEQID: 16 unclaimed DNA 314783-35-2 314783-36-3 314783-37-4 314783-38-5 314783-39-6 314783-40-9 314783-41-0 314783-42-1 314783-43-2 314783-44-3 314783-45-4 314783-46-5 314783-47-6 314783-48-7 314783-49-8
RL: PRP (Properties)
(unclaimed nucleotide sequence; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

IT 314783-27-2, 1: PN: US6165745 SEQID: 9 unclaimed DNA 314783-28-3, 2: PN: US6165745 SEQID: 10 unclaimed DNA 314783-29-4, 3: PN: US6165745 SEQID: 11 unclaimed DNA 314783-30-7, 4: PN: US6165745 SEQID: 12 unclaimed DNA 314783-31-8, 5: PN: US6165745 SEQID: 13 unclaimed DNA 314783-32-9, 6: PN: US6165745 SEQID: 14 unclaimed DNA 314783-33-0, 7: PN: US6165745 SEQID: 15 unclaimed DNA 314783-34-1, 8: PN: US6165745 SEQID: 16 unclaimed DNA 314783-35-2 314783-36-3 314783-37-4 314783-38-5 314783-39-6 314783-40-9 314783-41-0 314783-42-1 314783-43-2 314783-44-3 314783-45-4 314783-46-5 314783-47-6 314783-48-7 314783-49-8
RL: PRP (Properties)
(unclaimed nucleotide sequence; recombinant production of Ig-like domains in prokaryotic cells for use in immunization)

RE.CNT 1 THERE ARE 1 CITED REFERENCES AVAILABLE FOR THIS RECORD
RE
(1) Waldmann; Science 1991, V252, P1657 MEDLINE

L28 ANSWER 4 OF 35 HCAPLUS COPYRIGHT 2004 ACS on STN
AN 1999:571804 HCAPLUS
DN 131:194304
ED Entered STN: 09 Sep 1999
TI Peptides and pharmaceutical compositions thereof for treatment of disorders or diseases associated with abnormal protein folding into amyloid or amyloid-like deposits
IN Soto-Jara, Claudio; Baumann, Marc H.; Frangione, Blas
PA New York University, USA
SO U.S., 32 pp., Cont.-in-part of U.S. Ser. No. 478,326.
CODEN: USXXAM
DT **Patent**
LA English
IC ICM A61K038-00
NCL 514014000
CC 1-12 (Pharmacology)
FAN.CNT 3

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE	
PI	US 5948763	A	19990907	US 1996-630645	19960410	<--
	CA 2222690	AA	19961219	CA 1996-2222690	19960606	<--
	WO 9639834	A1	19961219	WO 1996-US10220	19960606	<--
	W: AU, CA, JP					
	RW: AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE					
	AU 9661129	A1	19961230	AU 1996-61129	19960606	<--
	AU 715662	B2	20000210			
	EP 843516	A1	19980527	EP 1996-918482	19960606	<--

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI

	JP 2001519753	T2	20011023	JP 1997-502245	19960606	<--
	US 6462171	B1	20021008	US 1996-766596	19961212	<--
	US 2003087407	A1	20030508	US 2002-235483	20020906	<--
PRAI	US 1995-478326	A2	19950607	<--		
	US 1996-630645	A	19960410			
	WO 1996-US10220	W	19960606			
	US 1996-766596	A1	19961212			

AB Peptides capable of interacting with a hydrophobic structural determinant on a protein or peptide for amyloid or amyloid-like deposit formation inhibit and structurally block the abnormal folding of proteins and peptides into amyloid or amyloid-like deposits. Methods for preventing, treating or detecting disorders or diseases associated with amyloid-like fibril deposits, such as Alzheimer's disease and prion-related encephalopathies, are also provided.

ST peptide pharmaceutical protein folding amyloid disorder; Alzheimer drug peptide protein folding; prion encephalopathy drug peptide protein folding

IT Amyloid
RL: BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
(A, and amyloid L; peptides for treatment of diseases associated with abnormal protein folding into amyloid or amyloid-like deposits)

IT Apolipoproteins
RL: BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
(A-I, amyloid; peptides for treatment of diseases associated with abnormal protein folding into amyloid or amyloid-like deposits)

IT Apolipoproteins
RL: BAC (Biological activity or effector, except adverse); BSU (Biological study, unclassified); BIOL (Biological study)
(E; peptides for treatment of diseases associated with abnormal protein folding into amyloid or amyloid-like deposits)

IT Proteins, specific or class
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(amphoterin, amyloid fragment derived from; peptides for treatment of diseases associated with abnormal protein folding into amyloid or amyloid-like deposits)

IT Gelsolin
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(amyloid; peptides for treatment of diseases associated with abnormal protein folding into amyloid or amyloid-like deposits)

IT Organelle
(fibril; peptides for treatment of diseases associated with abnormal protein folding into amyloid or amyloid-like deposits)

IT Amyloidosis
Anti-Alzheimer's agents
Cytoprotective agents
Molecular association
Protein folding
β-Sheet
(peptides for treatment of diseases associated with abnormal protein folding into amyloid or amyloid-like deposits)

IT Peptides, biological studies
RL: BAC (Biological activity or effector, except adverse); BPR (Biological process); BSU (Biological study, unclassified); THU (Therapeutic use); BIOL (Biological study); PROC (Process); USES (Uses)
(peptides for treatment of diseases associated with abnormal protein folding into amyloid or amyloid-like deposits)

IT Amyloid

Prion **proteins**
RL: **BPR (Biological process)**; BSU (Biological study, unclassified); BIOL (Biological study); **PROC (Process)**
(peptides for treatment of diseases associated with abnormal protein folding into amyloid or amyloid-like deposits)

IT Conformation
Secondary structure
(protein; peptides for treatment of diseases associated with abnormal protein folding into amyloid or amyloid-like deposits)

IT Amyloid
RL: BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
(β-; peptides for treatment of diseases associated with abnormal protein folding into amyloid or amyloid-like deposits)

IT Microglobulins
RL: BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
(β2-, amyloid; peptides for treatment of diseases associated with abnormal protein folding into amyloid or amyloid-like deposits)

IT Amino acids, biological studies
RL: BOC (Biological occurrence); BSU (Biological study, unclassified); BIOL (Biological study); OCCU (Occurrence)
(D-; peptides for treatment of diseases associated with abnormal protein folding into amyloid or amyloid-like deposits)

IT 164257-32-3 182912-72-7 182912-74-9 182912-76-1 182912-80-7
186606-60-0 186606-70-2 186606-72-4 186606-88-2 186606-93-9
186606-96-2 186607-00-1 186607-08-9 186607-12-5 242125-68-4
242125-70-8
RL: BAC (Biological activity or effector, except adverse); BSU (Biological study, unclassified); PRP (Properties); THU (Therapeutic use); BIOL (Biological study); USES (Uses)
(peptides for treatment of diseases associated with abnormal protein folding into amyloid or amyloid-like deposits)

IT 147-85-3, Proline, biological studies
RL: BOC (Biological occurrence); BSU (Biological study, unclassified); BIOL (Biological study); OCCU (Occurrence)
(peptides for treatment of diseases associated with abnormal protein folding into amyloid or amyloid-like deposits)

IT 186606-80-4 186606-84-8 242125-69-5
RL: BPR (Biological process); BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study); PROC (Process)
(peptides for treatment of diseases associated with abnormal protein folding into amyloid or amyloid-like deposits)

IT 182912-66-9 186606-30-4 186606-34-8 186606-39-3 186606-43-9
186606-48-4 186606-54-2
RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
(peptides for treatment of diseases associated with abnormal protein folding into amyloid or amyloid-like deposits)

RE.CNT 14 THERE ARE 14 CITED REFERENCES AVAILABLE FOR THIS RECORD
RE

(1) Anon; WO 9628471 1996 HCAPLUS
(2) Anon; WO 9721728 1997 HCAPLUS
(3) Borman; Science 1996, P33 HCAPLUS
(4) Burgess; The Journal of Cell Biology 1990, V111, P2129 HCAPLUS
(5) Chou; Ann Rev Biochem 1978, V47, P251 HCAPLUS
(6) Hilbich; J Mol Biol 1992, V228, P460 HCAPLUS
(7) Lazar; Molecular and Cellular Biology 1988, V8(3), P1247 HCAPLUS
(8) Rudinger; Peptide Hormones 1976, P6
(9) Soto, C; Journal of Biochemistry 1995, V20(7), P3063

(10) Soto, C; Neuroscience Letters 1995, V186, P115 HCAPLUS
(11) Soto, C; Structural Determinants of the Alzheimer's Amyloid β-Peptide Journal of Neurochemistry 1994, V63, P1191 HCAPLUS
(12) Wille; Ciba Foundation Symposium 199 1996, P181 HCAPLUS
(13) Wood; Biochemistry 1995, V34(3), P724 HCAPLUS
(14) Wood, S; Biochemistry 1995, V34, P724 HCAPLUS

L28 ANSWER 6 OF 35 HCAPLUS COPYRIGHT 2004 ACS on STN
AN 1998:293637 HCAPLUS
DN 128:318013
ED Entered STN: 20 May 1998
TI Process for bacterial production of polypeptides using DsbA and DsbC, for improved folding and disulfide bonding, and a mutant PstS, allowing de-repression at higher phosphate levels
IN Joly, John C.; Swartz, James R.
PA Genentech, Inc., USA
SO PCT Int. Appl., 45 pp.
CODEN: PIXXD2
DT **Patent**
LA English
IC ICM C12N015-70
ICS C07K014-245; C07K014-65
CC 3-2 (Biochemical Genetics)
Section cross-reference(s): 10, 16
FAN.CNT 3

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	WO 9818946	A1	19980507	WO 1997-US18383	19971009
	W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
	RW: GH, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
	US 5789199	A	19980804	US 1996-741727	19961031 <--
	AU 9748163	A1	19980522	AU 1997-48163	19971009
PRAI	US 1996-741727	A	19961031		
	US 1994-333912	A2	19941103 <--		
	WO 1997-US18383	W	19971009		

AB Claimed is a process for producing a heterologous polypeptide in bacteria comprising culturing bacterial cells lacking native pstS gene and containing: a pstS gene encoding a variant with a substitution in the phosphate-binding region; genes dsbA and dsbC; the gene encoding the heterologous polypeptide; a signal sequence for secretion of DsbA, DsbC, and the heterologous peptide; an inducible promoter for the dsbA or dsbC gene; an alkaline phosphatase promoter for the heterologous polypeptide gene; and the native pstS promoter for control of the variant pstS gene. The culturing take place under conditions where expression of DsbA or DsbC is induced prior to induction of the heterologous polypeptide, with an inorg. phosphate concentration in the medium above the cells' starvation level, and the heterologous polypeptide is secreted into the periplasm along with DsbA or DsbC, or is secreted into the medium. The mutation in the PstS phosphate-binding protein, normally a repressor protein, decreases the phosphate affinity, allowing polypeptide induction by the bacterial host at phosphate concns. higher than the starvation level. Specific substitution variants of the pstS nucleic acid and protein are also claimed.

ST pstS dsbA dsbC protein cloning Escherichia; phosphate derepression pstS cloning enterobacteriaceae; substitution mutation pstS cloning Escherichia

IT Promoter (genetic element)
RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(alkaline phosphatase; bacterial production of polypeptides using DsbA and DsbC, for improved folding and disulfide bonding, and a mutant PstS, allowing de-repression at higher phosphate levels)

IT Culture media
Disulfide group
Enterobacteriaceae
Escherichia coli
Mammal (Mammalia)
Molecular cloning
Plasmid vectors
Protein folding
Transcriptional regulation
(bacterial production of polypeptides using DsbA and DsbC, for improved folding and disulfide bonding, and a mutant PstS, allowing de-repression at higher phosphate levels)

IT **Proteins, general, preparation**
RL: **BPN (Biosynthetic preparation)**; BIOL (Biological study); **PREP (Preparation)**
(bacterial production of polypeptides using DsbA and DsbC, for improved folding and disulfide bonding, and a mutant PstS, allowing de-repression at higher phosphate levels)

IT Gene, microbial
RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(dsbA; bacterial production of polypeptides using DsbA and DsbC, for improved folding and disulfide bonding, and a mutant PstS, allowing de-repression at higher phosphate levels)

IT Gene, microbial
RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(dsbC; bacterial production of polypeptides using DsbA and DsbC, for improved folding and disulfide bonding, and a mutant PstS, allowing de-repression at higher phosphate levels)

IT **Proteins, specific or class**
RL: **BPR (Biological process)**; BSU (Biological study, unclassified); BUU (Biological use, unclassified); BIOL (Biological study); **PROC (Process)**; USES (Uses)
(gene DsbA; bacterial production of polypeptides using DsbA and DsbC, for improved folding and disulfide bonding, and a mutant PstS, allowing de-repression at higher phosphate levels)

IT **Proteins, specific or class**
RL: **BPR (Biological process)**; BSU (Biological study, unclassified); BUU (Biological use, unclassified); BIOL (Biological study); **PROC (Process)**; USES (Uses)
(gene dsbC; bacterial production of polypeptides using DsbA and DsbC, for improved folding and disulfide bonding, and a mutant PstS, allowing de-repression at higher phosphate levels)

IT Promoter (genetic element)
RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(inducible; bacterial production of polypeptides using DsbA and DsbC, for improved folding and disulfide bonding, and a mutant PstS, allowing de-repression at higher phosphate levels)

IT Organelle
(periplasm; bacterial production of polypeptides using DsbA and DsbC, for

improved folding and disulfide bonding, and a mutant PstS, allowing de-repression at higher phosphate levels)

IT **Proteins, specific or class**
RL: **BPR (Biological process)**; BSU (Biological study, unclassified); BUU (Biological use, unclassified); BIOL (Biological study); **PROC (Process)**; USES (Uses)
(phosphate-binding, gene pstS; bacterial production of polypeptides using DsbA and DsbC, for improved folding and disulfide bonding, and a mutant PstS, allowing de-repression at higher phosphate levels)

IT Secretion (process)
(protein; bacterial production of polypeptides using DsbA and DsbC, for improved folding and disulfide bonding, and a mutant PstS, allowing de-repression at higher phosphate levels)

IT Promoter (genetic element)
RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(pstS; bacterial production of polypeptides using DsbA and DsbC, for improved folding and disulfide bonding, and a mutant PstS, allowing de-repression at higher phosphate levels)

IT Gene, microbial
RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(pstS; substitution variants; bacterial production of polypeptides using DsbA and DsbC, for improved folding and disulfide bonding, and a mutant PstS, allowing de-repression at higher phosphate levels)

IT Transcription factors
RL: BPR (Biological process); BSU (Biological study, unclassified); BUU (Biological use, unclassified); BIOL (Biological study); PROC (Process); USES (Uses)
(repressors, gene pstS; bacterial production of polypeptides using DsbA and DsbC, for improved folding and disulfide bonding, and a mutant PstS, allowing de-repression at higher phosphate levels)

IT Mutation
(substitution; bacterial production of polypeptides using DsbA and DsbC, for improved folding and disulfide bonding, and a mutant PstS, allowing de-repression at higher phosphate levels)

IT 14265-44-2, Phosphate, biological studies
RL: BAC (Biological activity or effector, except adverse); BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
(bacterial production of polypeptides using DsbA and DsbC, for improved folding and disulfide bonding, and a mutant PstS, allowing de-repression at higher phosphate levels)

IT 14265-44-2, Phosphate, biological studies
RL: BAC (Biological activity or effector, except adverse); BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
(bacterial production of polypeptides using DsbA and DsbC, for improved folding and disulfide bonding, and a mutant PstS, allowing de-repression at higher phosphate levels)

RE.CNT 2 THERE ARE 2 CITED REFERENCES AVAILABLE FOR THIS RECORD
RE
(1) Bass, S; US 5304472 A 1994 HCAPLUS
(2) Genentech Inc; WO 9614422 A 1996 HCAPLUS

L28 ANSWER 7 OF 35 HCAPLUS COPYRIGHT 2004 ACS on STN
AN 1998:219824 HCAPLUS
DN 128:280594
ED Entered STN: 18 Apr 1998
TI Method of purifying protein from inclusion bodies

IN Li, Yuling; Oelkuct, Mark; Gentz, Reiner L.
PA Human Genome Sciences, Inc., USA
SO PCT Int. Appl., 104 pp.
CODEN: PIXXD2
DT **Patent**
LA English
IC ICM C07K001-113
ICS C07K001-36; C07K014-50; C07K014-52
CC 9-16 (Biochemical Methods)
Section cross-reference(s): 15, 16
FAN.CNT 10

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	WO 9814467	A1	19980409	WO 1997-US17510	19970930
	W: CA, JP				
	RW: AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
	US 6001606	A	19991214	US 1996-722719	19960930 <--
	US 5912327	A	19990615	US 1997-821637	19970320
	EP 975657	A1	20000202	EP 1997-944561	19970930
	R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
	JP 2001503614	T2	20010321	JP 1998-516755	19970930
PRAI	US 1996-722719	A	19960930		
	US 1996-722723	A	19960930		
	US 1997-821637	A	19970320		
	US 1994-208339	A2	19940308 <--		
	US 1995-446881	B2	19950505 <--		
	US 1995-465682	B2	19950606 <--		
	US 1995-468775	B2	19950606 <--		
	US 1995-4517P	P	19950929 <--		
	WO 1996-US15592	A2	19960927		
	WO 1997-US17510	W	19970930		

AB The present invention relates to processes for the purification of proteins. More specifically, methods for solubilizing and purifying proteins expressed in an insol. form using low concns. of chaotropic agents, such as guanidine salts, are provided. Also provided are methods for refolding proteins solubilized according to the present invention.

ST purifying protein inclusion body

IT **Proteins, specific or class**
RL: **PUR (Purification or recovery); PREP (Preparation)**
(Ck-α-4; method of purifying protein from inclusion bodies)

IT **Proteins, specific or class**
RL: **PUR (Purification or recovery); PREP (Preparation)**
(Ck-β-13; method of purifying protein from inclusion bodies)

IT **Proteins, specific or class**
RL: **PUR (Purification or recovery); PREP (Preparation)**
(FGF-13; method of purifying protein from inclusion bodies)

IT **Proteins, specific or class**
RL: **PUR (Purification or recovery); PREP (Preparation)**
(M-CIF; method of purifying protein from inclusion bodies)

IT **Proteins, specific or class**
RL: **PUR (Purification or recovery); PREP (Preparation)**
(MIP-1α; method of purifying protein from inclusion bodies)

IT **Proteins, specific or class**
RL: **PUR (Purification or recovery); PREP (Preparation)**
(MIP-4; method of purifying protein from inclusion bodies)

IT **Proteins, specific or class**
RL: **PUR (Purification or recovery); PREP (Preparation)**
(MPIF-1; method of purifying protein from inclusion bodies)

IT **Proteins, specific or class**

RL: **PUR (Purification or recovery)**; **PREP (Preparation)**
(MPIF-1d23; method of purifying protein from inclusion bodies)
IT Denaturants
(chaotropic; method of purifying protein from inclusion bodies)
IT Toxins
RL: PUR (Purification or recovery); PREP (Preparation)
(endotoxins; method of purifying protein from inclusion bodies)
IT Animal cell
(mammalian; method of purifying protein from inclusion bodies)
IT Bacteria (Eubacteria)
Inclusion bodies
Insect (Insecta)
Liquid chromatography
Microorganism
Protein folding
Purification
Solubilization
Ultrafiltration
Yeast
(method of purifying protein from inclusion bodies)
IT Chemokines
Proteins, general, preparation
RL: **PUR (Purification or recovery)**; **PREP (Preparation)**
(method of purifying protein from inclusion bodies)
IT Filtration
(microfiltration; method of purifying protein from inclusion bodies)
IT Chromatography
(tandem; method of purifying protein from inclusion bodies)
IT 50-01-1, Guanidine hydrochloride 57-13-6, Urea, uses 113-00-8D, Guanidine, salts 35754-33-7
RL: NUU (Other use, unclassified); USES (Uses)
(method of purifying protein from inclusion bodies)
RE.CNT 11 THERE ARE 11 CITED REFERENCES AVAILABLE FOR THIS RECORD
RE
(1) American Cyanamid; EP 0432419 A 1991 HCAPLUS
(2) Hart, R; BIOTECHNOLOGY AND BIOENGINEERING INCLUDING: SYMPOSIUM BIOTECHNOLOGY IN ENERGY PRODUCTION AND CONSERVATION 1992, V39(11), P1112 HCAPLUS
(3) Human Genome Sciences; WO 9517092 A 1995 HCAPLUS
(4) Human Genome Sciences; WO 9634891 A 1996 HCAPLUS
(5) Human Genome Sciences; WO 9639508 A 1996 HCAPLUS
(6) Human Genome Sciences; WO 9715594 A 1997 HCAPLUS
(7) International Mineral And Chemical Corporation; EP 0215625 A 1987 HCAPLUS
(8) Mikulski, A; CURR CHEM INFECT DIS, PROC INT CONGR CHEMOTHER, 11TH (MEETING DATE 1979) 1980, V2, P11746 HCAPLUS
(9) Patel, V; J EXP MED 1997, V185(7), P1163 HCAPLUS
(10) Proost, P; METHODS 1996, V10(1), P82 HCAPLUS
(11) Tsuji, T; BIOCHEMISTRY 1991, V26, P3129

L28 ANSWER 8 OF 35 HCAPLUS COPYRIGHT 2004 ACS on STN
AN 1998:816063 HCAPLUS
DN 130:62019
ED Entered STN: 01 Jan 1999
TI Method of expressing antifreeze proteins in yeast
IN Tripp, Matthew; Lusk, Lance; Rhodes, Thomas; Huige, Nick; Kot, Edward; Chicoye, Etzer; Barney, Michael C.; Bower, Patricia A.; Cronan, Charles L.
PA Miller Brewing Company, USA
SO U.S., 25 pp., Cont. of U.S. Ser. No. 917,216, abandoned.
CODEN: USXXAM
DT **Patent**

LA English
IC ICM C12P021-02
ICS C12N015-81; C12N001-19
NCL 435069700
CC 3-2 (Biochemical Genetics)
Section cross-reference(s): 10
FAN.CNT 1

	PATENT NO.	KIND	DATE		APPLICATION NO.	DATE	
PI	US 5849537	A	19981215		US 1994-180524	19940112	<--
	US 5928877	A	19990727		US 1997-975166	19971120	<--
PRAI	US 1989-409217		19890919	<--			
	US 1990-486333		19900228	<--			
	US 1992-917216		19920720	<--			
	US 1994-180524		19940112	<--			

AB Yeast is genetically engineered by transformation with an expression vector containing a natural yeast secretion signal sequence combined appropriately with a chemical synthesized gene encoding antifreeze protein resulting in the expression, proper processing, and secretion of antifreeze protein which is heterologous to yeast in recoverable amts. Disclosed are DNA sequences comprising structural genes encoding peptides having amino acid sequences with the biochem. or physiochem. properties of antifreeze protein and a method of combining the antifreeze protein gene sequences with appropriate expression vectors.

ST Saccharomyces antifreeze protein expression secretion processing

IT **Proteins, specific or class**
RL: **BPN (Biosynthetic preparation)**; **BPR (Biological process)**; BSU (Biological study, unclassified); BIOL (Biological study); **PREP (Preparation)**; **PROC (Process)**
(antifreeze; method of expressing antifreeze proteins in yeast)

IT Food industry
Ice cream
(applications in food industry; method of expressing antifreeze proteins in yeast)

IT Chimeric gene
RL: BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation)
(encoding secretion leader and AFP sequences and junction/processing peptide; method of expressing antifreeze proteins in yeast)

IT Post-translational processing
Protein folding
(generation of proteins correctly processed and secreted and folded; method of expressing antifreeze proteins in yeast)

IT Pseudopleuronectes americanus
(high similarity to sequences from; method of expressing antifreeze proteins in yeast)

IT Concentration (process)
(method for protein concentration; method of expressing antifreeze proteins in yeast)

IT Protein engineering
Saccharomyces cerevisiae
(method of expressing antifreeze proteins in yeast)

IT Secretion (process)
(protein, generation of proteins correctly processed and secreted and folded; method of expressing antifreeze proteins in yeast)

IT Fermentation
Membrane filters
Ultrafilters
(secreted protein recovery by passing fermented broth through filter

and then through ultrafiltration membrane; method of expressing antifreeze proteins in yeast)

IT 217824-35-6P 217824-36-7P
RL: BPN (Biosynthetic preparation); BSU (Biological study, unclassified); IMF (Industrial manufacture); PRP (Properties); BIOL (Biological study); PREP (Preparation)
(amino acid sequence; method of expressing antifreeze proteins in yeast)

RE.CNT 16 THERE ARE 16 CITED REFERENCES AVAILABLE FOR THIS RECORD

RE
(1) Anon; WO 00876 1988
(2) Anon; WO 88/07076 1988 HCAPLUS
(3) Brake; PNAS 1984, V81, P4642 HCAPLUS
(4) Britz; Austral J Biotech 1987, V1(3), P29 HCAPLUS
(5) Davies; PNAS 1982, V79, P335 HCAPLUS
(6) Davies; PNAS 1982, V79, P335 HCAPLUS
(7) Ginsberg; J Clin Endocrin Metab 1979, V48(1), P43 HCAPLUS
(8) Gourlie; J Biol Chem 1984, V259, P14960 HCAPLUS
(9) Gronlic; J Biol Chem 1984, V259, P14960
(10) Kawasaki; US 4839283 1989 HCAPLUS
(11) Murray; US 4769328 1988 HCAPLUS
(12) Pesole; Nuc Acid Res 1988, V16, P1715 HCAPLUS
(13) Pesole; Nuc Acid Res 1988, P1715 HCAPLUS
(14) Peterson; A Biological Antifreeze 1986, V130, P330
(15) Scopes; Protein Purification 1982, P183
(16) Warren; US 5118792 1992 HCAPLUS

L28 ANSWER 9 OF 35 HCAPLUS COPYRIGHT 2004 ACS on STN
AN 1998:604690 HCAPLUS
DN 129:226609
ED Entered STN: 24 Sep 1998
TI Refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion bodies
IN Builder, Stuart; Hart, Roger; Lester, Philip; Reifsnyder, David
PA Genentech, Inc., USA
SO U.S., 38 pp., Cont.-in-part of U.S. Ser. No. 110,664.
CODEN: USXXAM
DT **Patent**
LA English
IC ICM C07K014-475
ICS A61K038-22
NCL 530399000
CC 3-1 (Biochemical Genetics)
Section cross-reference(s): 6

FAN.CNT 2

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE	
PI	US 5808006	A	19980915	US 1994-318628	19941011	<--
	US 5663304	A	19970902	US 1993-110664	19930820	<--
	WO 9506064	A1	19950302	WO 1994-US9120	19940815	<--
	W: CA, JP, US					
	RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE					
PRAI	US 1993-110664	A2	19930820	<--		
	WO 1994-US9120	W	19940815	<--		

AB A method for solubilizing and refolding peptides manufactured in bacterial hosts and accumulated as inclusion bodies is described. The method is optimized for the recovery of insulin-like growth factor and its analogs and is inexpensive. The polypeptide is resuspended at 0.1-15 mg/mL in a buffer having a pH of about 7-12 of 5-40 volume/volume% of an alc. or polar aprotic solvent, about 0.2-3M of an alkaline earth, alkali metal, or ammonium

salt, about 0.1-9M of a chaotropic agent, and about 0.10-15 μM of a copper or manganese salt. The protein is allowed to refold by incubating it in this buffer. The presence of the low concns. of copper or manganese minimizes the formation of incorrectly folded proteins and avoids the need for disulfide exchange agents. The method can also be used in two-phase systems where cell lysates are fractionated by phase partition and the phase containing the inclusion bodies is under conditions suitable for solubilization and renaturation. The method is demonstrated with IGF-1 manufactured in Escherichia coli by expression of a cDNA. From a large-scale fermentation (600-800 L) the protein could be refolded with a recovery of .apprx.50%. Expts. using two-phase systems are reported.

ST inclusion body protein solubilization renaturation

IT Polar solvents
(aprotic, as solvent in solubilization and renaturation medium; refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion bodies)

IT Alcohols, processes
RL: PEP (Physical, engineering or chemical process); PROC (Process)
(as solvent in solubilization and renaturation medium; refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion bodies)

IT Denaturants
(chaotropic, refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion bodies)

IT Neurotrophic factors
Proteins, general, preparation
RL: **BMF (Bioindustrial manufacture)**; **BPN (Biosynthetic preparation)**; BIOL (Biological study); **PREP (Preparation)**
(manufactured as inclusion bodies; refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion bodies)

IT Solutes
(osmolytes, refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion bodies)

IT Fermentation
(protein; refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion bodies)

IT Buffers
Inclusion bodies
Protein folding
Renaturation
(refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion bodies)

IT Alkali metal compounds
Alkaline earth compounds
RL: PEP (Physical, engineering or chemical process); PROC (Process)
(salts, refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion bodies)

IT 56-40-6, Glycine, processes 1135-40-6, CAPS 73463-39-5, CAPSO
RL: PEP (Physical, engineering or chemical process); PROC (Process)
(as buffer; refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion bodies)

IT 57-13-6, Urea, processes
RL: PEP (Physical, engineering or chemical process); PROC (Process)
(as chaotropic agent; refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion bodies)

IT 56-81-5, Glycerol, processes
RL: PEP (Physical, engineering or chemical process); PROC (Process)
(as solvent and osmolyte; refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion

bodies)
IT 57-55-6, Propylene glycol, processes 64-17-5, Ethanol, processes 67-56-1, Methanol, processes 67-63-0, Iso-propanol, processes 67-68-5, DMSO, processes 68-12-2, Dimethylformamide, processes 71-23-8, n-Propanol, processes 75-05-8, Acetonitrile, processes 75-65-0, Tert-Butanol, processes 109-99-9, Tetrahydrofuran, processes 123-91-1, Dioxane, processes 872-50-4, N-Methylpyrrolidone, processes
RL: PEP (Physical, engineering or chemical process); PROC (Process)
(as solvent; refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion bodies)
IT 9002-72-6P, Growth hormone
RL: BMF (Bioindustrial manufacture); BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation)
(manufactured as inclusion bodies; refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion bodies)
IT 57-50-1, Sucrose, processes
RL: PEP (Physical, engineering or chemical process); PROC (Process)
(osmolyte; refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion bodies)
IT 52-90-4, Cysteine, processes 3483-12-3, Dithiothreitol 7439-96-5D, Manganese, salts, processes 7440-50-8D, Copper, salts, processes 7447-39-4, Copper chloride, processes 7758-98-7, Copper sulfate, processes 7773-01-5, Manganese chloride 7785-87-7, Manganese sulfate
RL: PEP (Physical, engineering or chemical process); PROC (Process)
(reducing agent; refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion bodies)
IT 61912-98-9P, Insulin-like growth factor 67763-96-6P, Insulin-like growth factor 1
RL: BMF (Bioindustrial manufacture); BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation)
(refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion bodies)
IT 7440-09-7D, Potassium, salts, processes 7440-23-5D, Sodium, salts, processes 7487-88-9, Magnesium sulfate, processes 7647-14-5, Sodium chloride, processes 7664-41-7D, Ammonia, salts, processes 7757-82-6, Sodium sulfate, processes 7783-20-2, Ammonium sulfate, processes 7786-30-3, Magnesium chloride, processes 12125-02-9, Ammonium chloride, processes
RL: PEP (Physical, engineering or chemical process); PROC (Process)
(refolding of improperly folded polypeptides like recombinant insulin-like growth factor recovered from inclusion bodies)
RE.CNT 112 THERE ARE 112 CITED REFERENCES AVAILABLE FOR THIS RECORD
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L28 ANSWER 13 OF 35 HCAPLUS COPYRIGHT 2004 ACS on STN
AN 1997:211221 HCAPLUS
DN 126:197114
ED Entered STN: 02 Apr 1997
TI Process for protein refolding by means of buffer exchange using a continuous stationary phase capable of separating proteins from salt
IN Seely, Robert J.; Ladisch, Michael R.
PA Amgen Inc., USA; Purdue Research Foundation
SO PCT Int. Appl., 26 pp.
CODEN: PIXXD2
DT **Patent**
LA English
IC ICM C07K001-113
ICS C07K001-14
CC 9-3 (Biochemical Methods)
Section cross-reference(s): 2, 6, 16

FAN.CNT 1

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE	
PI	WO 9704003	A2	19970206	WO 1996-US11581	19960711	<--
	WO 9704003	A3	19970227			
	W: AL, AM, AT, AU, AZ, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EE, ES, FI, GB, GE, HU, IL, IS, JP, KE, KG, KP, KR, KZ, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG					
	RW: KE, LS, MW, SD, SZ, UG, AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN					
	AU 9665444	A1	19970218	AU 1996-65444	19960711	<--
	AU 707719	B2	19990715			
	EP 840745	A2	19980513	EP 1996-925295	19960711	<--
	R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI					
	JP 11509551	T2	19990824	JP 1996-506744	19960711	<--
	CA 2227318	C	20010925	CA 1996-2227318	19960711	<--
	ZA 9606151	A	19970204	ZA 1996-6151	19960719	<--
PRAI	US 1995-505420	A	19950721	<--		
	WO 1996-US11581	W	19960711			

AB Disclosed is a quick and efficient refolding process which uses size exclusion chromatog. with a cellulosic rolled stationary phase to sep. rapidly the reduced, denatured protein from the denaturant solution, thereby promoting high protein refold yields at higher protein concns., while significantly decreasing the volume needed to achieve protein refolding.

ST protein refolding buffer exchange stationary phase; size exclusion chromatog protein refolding; cellulose rolled stationary phase protein refolding; renaturation protein rolled stationary phase; fermn recombinant protein prodn refolding

IT Neurotrophic factors
RL: PEP (Physical, engineering or chemical process); PRP (Properties); PROC (Process)
(brain-derived; protein refolding by buffer exchange using continuous stationary phase that seps. proteins from salt)

IT Textiles
(cotton-polyester, DEAE-derivatized; protein refolding by buffer exchange using continuous stationary phase that seps. proteins from salt)

IT Salts, processes
RL: REM (Removal or disposal); PROC (Process)
(desalting; protein refolding by buffer exchange using continuous stationary phase that seps. proteins from salt)

IT Neurotrophic factors
RL: PEP (Physical, engineering or chemical process); PRP (Properties); PROC (Process)
(glial-derived; protein refolding by buffer exchange using continuous stationary phase that seps. proteins from salt)

IT Buffers
Denaturants
Fermentation
Renaturation
(protein refolding by buffer exchange using continuous stationary phase that seps. proteins from salt)

IT **Proteins, general, properties**
RL: **PEP (Physical, engineering or chemical process)**; PRP (Properties); **PROC (Process)**
(protein refolding by buffer exchange using continuous stationary phase that seps. proteins from salt)

IT **Protein folding**

(refolding; protein refolding by buffer exchange using continuous stationary phase that seps. proteins from salt)
IT Size-exclusion chromatography
(stationary phases; protein refolding by buffer exchange using continuous stationary phase that seps. proteins from salt)
IT 50-01-1, Guanidine hydrochloride
RL: BUU (Biological use, unclassified); PEP (Physical, engineering or chemical process); REM (Removal or disposal); BIOL (Biological study); PROC (Process); USES (Uses)
(protein refolding by buffer exchange using continuous stationary phase that seps. proteins from salt)
IT 9004-34-6, Cellulose, uses
RL: NUU (Other use, unclassified); USES (Uses)
(protein refolding by buffer exchange using continuous stationary phase that seps. proteins from salt)
IT 9061-61-4, Nerve growth factor 122320-05-2, Secretory leukocyte protease inhibitor 130939-66-1, Neurotrophin 3
RL: PEP (Physical, engineering or chemical process); PRP (Properties); PROC (Process)
(protein refolding by buffer exchange using continuous stationary phase that seps. proteins from salt)

L28 ANSWER 15 OF 35 HCAPLUS COPYRIGHT 2004 ACS on STN
AN 2003:665844 HCAPLUS
DN 139:178810
ED Entered STN: 27 Aug 2003
TI A novel method of production of correctly folded insulin
IN Srinivasa, Bachally Ramasastry; Ramachandran, Janakiraman
PA Astra Research Centre India, India
SO Indian, 26 pp.
CODEN: INXXAP
DT **Patent**
LA English
IC ICM C12P021-00
CC 16-2 (Fermentation and Bioindustrial Chemistry)
Section cross-reference(s): 2, 3
FAN.CNT 1

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	IN 179830	A	19971220	IN 1994-MA1197	19941202 <--
PRAI	IN 1994-MA1197		19941202 <--		

AB Disclosed herein is a novel method of production of correctly folded insulin which comprises the steps of (a) Constructing a plasmid that encodes a GST-Met-B chain of insulin--Met--Met--A chain of insulin fusion protein (proinsulin): (b) Transforming the plasmid obtained in step (a) into a suitable E.coli Strain such as herein described: (c) Expressing the fusion product (proinsulin) by culturing the bacteria by known methods; (d) Isolating the fusion product (proinsulin) by known methods; (e) Cleaving the fusion product (proinsulin) to sep. GST and the mature insulin by known methods; (f) Cleaving the homoserine residues by a method such as herein described; and (g) Purifying the Insulin by ion exchange chromotog.
ST human insulin fusion protein folding sequence
IT Synthetic gene
RL: BPN (Biosynthetic preparation); BSU (Biological study, unclassified); BIOL (Biological study); PREP (Preparation)
(for human insulin; novel method of production of correctly folded insulin)
IT Gene, animal
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(for human insulin; novel method of production of correctly folded insulin)
IT Human

(genes and proteins; novel method of production of correctly folded insulin)
IT Fermentation
Genetic engineering
Ion exchange chromatography
Plasmid vectors
Protein folding
(novel method of production of correctly folded insulin)
IT Fusion **proteins** (chimeric **proteins**)
RL: **BCP (Biochemical process)**; **BMF (Bioindustrial manufacture)**; **BPN (Biosynthetic preparation)**; **PUR (Purification or recovery)**; RCT (Reactant); BIOL (Biological study); **PREP (Preparation)**; **PROC (Process)**; RACT (Reactant or reagent)
(novel method of production of correctly folded insulin)
IT Escherichia coli
(recombinant; novel method of production of correctly folded insulin)
IT 577797-80-9
RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
(amino acid sequence; novel method of production of correctly folded insulin)
IT 9004-10-8DP, Insulin, fusion protein with glutathione S-transferase
50812-37-8DP, Glutathione s transferase, fusion protein with proinsulin
RL: BCP (Biochemical process); BMF (Bioindustrial manufacture); BPN (Biosynthetic preparation); PUR (Purification or recovery); RCT (Reactant); BIOL (Biological study); PREP (Preparation); PROC (Process); RACT (Reactant or reagent)
(novel method of production of correctly folded insulin)
IT 9031-98-5, Carboxypeptidase
RL: BCP (Biochemical process); CAT (Catalyst use); BIOL (Biological study); PROC (Process); USES (Uses)
(novel method of production of correctly folded insulin)
IT 9004-10-8P, Insulin, biological studies
RL: BMF (Bioindustrial manufacture); BSU (Biological study, unclassified); BIOL (Biological study); PREP (Preparation)
(novel method of production of correctly folded insulin)
IT 506-68-3, Cyanogen bromide
RL: RCT (Reactant); RACT (Reactant or reagent)
(novel method of production of correctly folded insulin)
IT 577797-79-6
RL: BSU (Biological study, unclassified); PRP (Properties); BIOL (Biological study)
(nucleotide sequence; novel method of production of correctly folded insulin)
IT 577798-91-5, 4: PN: IN179830 PAGE: 11 unclaimed DNA
RL: PRP (Properties)
(unclaimed nucleotide sequence; novel method of production of correctly folded insulin)
IT 577706-44-6 577706-46-8 577798-92-6 577798-93-7 577800-86-3
RL: PRP (Properties)
(unclaimed sequence; novel method of production of correctly folded insulin)

L28 ANSWER 16 OF 35 HCAPLUS COPYRIGHT 2004 ACS on STN
AN 1997:397230 HCAPLUS
DN 127:13443
ED Entered STN: 27 Jun 1997
TI A screening method depending on protein folding for identifying potential pharmaceutical ligands for target proteins

IN Pakula, Andrew; Bowie, James
PA Scriptgen Pharmaceuticals, Inc., USA
SO Eur. Pat. Appl., 32 pp.
CODEN: EPXXDW
DT **Patent**
LA English
IC ICM G01N033-68
ICS G01N033-543
ICA C12Q001-37
CC 1-1 (Pharmacology)
FAN.CNT 3

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE	
PI	EP 770876	A1	19970502	EP 1996-610042	19961017	<--
	EP 770876	B1	20010418			
	R: AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LI, LU, MC, NL, PT, SE					
	CA 2184195	AA	19970426	CA 1996-2184195	19960826	<--
	CA 2184195	C	20020416			
	AU 9664298	A1	19970501	AU 1996-64298	19960828	<--
	AU 698862	B2	19981112			
	IL 119149	A1	20020310	IL 1996-119149	19960828	<--
	JP 09178746	A2	19970711	JP 1996-239252	19960910	<--
	JP 2952848	B2	19990927			
	BR 9604352	A	19980616	BR 1996-4352	19961004	<--
	AT 200579	E	20010415	AT 1996-610042	19961017	<--
	ES 2158269	T3	20010901	ES 1996-610042	19961017	<--
	PT 770876	T	20010928	PT 1996-610042	19961017	<--
	GR 3036138	T3	20010928	GR 2001-400992	20010627	<--
PRAI	US 1995-547889	A	19951025	<--		

AB A method for screening chemical compds. (test ligands) for potential pharmaceutical effectiveness is provided. The method identifies possible therapeutic test ligands by placing them in the presence of target proteins and determining their ability to increase or decrease the ratio of folded target protein to unfolded target protein. The present methods do not require that biochem. function of the target protein be known, nor that any other ligands be previously identified. The methodol. of the invention was used to identify ligands. e.g. inhibiting Hb S polymerization

ST protein folding therapeutic ligand screening; pharmaceutical ligand screening protein folding; Hb S polymn inhibitor screening

IT Rev **protein**
RL: **BPR (Biological process)**; BSU (Biological study, unclassified); BIOL (Biological study); **PROC (Process)**
(HIV; protein-folding method for identifying potential pharmaceutical ligands for target proteins)

IT Polymerization
(HbS, inhibitors; protein-folding method for identifying potential pharmaceutical ligands for target proteins)

IT Human immunodeficiency virus
(Rev protein; protein-folding method for identifying potential pharmaceutical ligands for target proteins)

IT Polyacrylamide gel electrophoresis
(denaturing; protein-folding method for identifying potential pharmaceutical ligands for target proteins)

IT Immunoassay
(enzyme-linked immunosorbent assay; protein-folding method for identifying potential pharmaceutical ligands for target proteins)

IT Conformation
(protein, target protein conformational domains; protein-folding method for identifying potential pharmaceutical ligands for target proteins)

IT Aggregation
Calorimetry
Circular dichroism spectroscopy
Denaturants
Detergents
Drug screening
Drugs
Fluorometry
Immobilization, biochemical
Immunoassay
Protein degradation
Protein folding
Temperature effects, biological
UV and visible spectroscopy
(protein-folding method for identifying potential pharmaceutical ligands for target proteins)

IT Ligands
RL: BAC (Biological activity or effector, except adverse); BPR (Biological process); BSU (Biological study, unclassified); THU (Therapeutic use); BIOL (Biological study); PROC (Process); USES (Uses)
(protein-folding method for identifying potential pharmaceutical ligands for target proteins)

IT Hemoglobins
Proteins, general, biological studies
RL: **BPR (Biological process)**; BSU (Biological study, unclassified); BIOL (Biological study); **PROC (Process)**
(protein-folding method for identifying potential pharmaceutical ligands for target proteins)

IT Amino acids, biological studies
RL: BSU (Biological study, unclassified); BIOL (Biological study)
(protein-folding method for identifying potential pharmaceutical ligands for target proteins)

IT Antibodies
RL: THU (Therapeutic use); BIOL (Biological study); USES (Uses)
(protein-folding method for identifying potential pharmaceutical ligands for target proteins)

IT Chaperonins
RL: THU (Therapeutic use); BIOL (Biological study); USES (Uses)
(protein-folding method for identifying potential pharmaceutical ligands for target proteins)

IT 9004-06-2, Elastase
RL: BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
(human neutrophil; protein-folding method for identifying potential pharmaceutical ligands for target proteins)

IT 138-81-8
RL: BAC (Biological activity or effector, except adverse); BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
(protein-folding method for identifying potential pharmaceutical ligands for target proteins)

IT 54-05-7, ST 121 58-93-5, Hydrochlorothiazide 59-66-5, Acetazolamide 63-74-1, Sulfanilamide 83-89-6, ST 439 960-57-6, ST 5196 972-18-9, ST 38624 1405-89-6, ST 56 1421-65-4, ST 41769 7149-45-3, ST 38904 7252-27-9, ST 16969 7252-50-8, ST 38473 13590-98-2, ST 39008 15190-13-3, ST 38775 23652-87-1, ST 41070 32022-06-3, ST 38626 37082-08-9, ST 38222 38714-92-0, ST 38218 50482-67-2, ST 39224 51798-45-9, Elastatinal 54978-84-6, ST 43883 149859-17-6, MDL 101146 190255-93-7, ST 9495 190256-96-3, ST 48775 190396-13-5, MDL 103900 190396-14-6, MDL 105373 190396-29-3, ST 69

RL: BAC (Biological activity or effector, except adverse); BPR (Biological process); BSU (Biological study, unclassified); THU (Therapeutic use); BIOL (Biological study); PROC (Process); USES (Uses)
(protein-folding method for identifying potential pharmaceutical ligands for target proteins)

IT 9001-03-0, Carbonic anhydrase 9002-03-3, Dihydrofolate reductase 9034-51-9, Hb A 9035-22-7, Hb S 50926-05-1
RL: BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
(protein-folding method for identifying potential pharmaceutical ligands for target proteins)

IT 53-57-6, NADPH 59-05-2, Methotrexate
RL: BPR (Biological process); BSU (Biological study, unclassified); THU (Therapeutic use); BIOL (Biological study); PROC (Process); USES (Uses)
(protein-folding method for identifying potential pharmaceutical ligands for target proteins)

IT 57-13-6, Urea, biological studies 9001-92-7, Protease 9073-78-3, Thermolysin 25215-10-5, Guanidinium 39450-01-6
RL: THU (Therapeutic use); BIOL (Biological study); USES (Uses)
(protein-folding method for identifying potential pharmaceutical ligands for target proteins)

L28 ANSWER 20 OF 35 WPIX COPYRIGHT 2004 THOMSON DERWENT on STN
AN 1997-087056 [08] WPIX
DNC C1997-028244
TI Aqueous formulation of tissue factor pathway inhibitor - contains charged polymer, e.g. dextran sulphate, to facilitate solubilisation, formulation purification and **refolding** of protein.
DC A96 B04 D16
IN ARVE, B H; BILD, G S; CHEN, B; DORIN, G J; GUSTAFSON, M E; HALENBECK, R F; HORA, M S; JOHNSON, G V; JOHNSON, K; MADANI, H; PATTISON, G L; RANA, R K; TSANG, M; HALLENBECK, R F; HOBA, M S
PA (CHIR) CHIRON CORP; (SEAR) SEARLE & CO G D
CYC 72
PI WO 9640784 A2 19961219 (199708)* EN 86 C07K014-81
RW: AT BE CH DE DK EA ES FI FR GB GR IE IT KE LS LU MC MW NL OA PT SD SE SZ UG
W: AL AM AT AU AZ BB BG BR BY CA CH CN CZ DE DK EE ES FI GB GE HU IL IS JP KE KG KP KR KZ LK LR LS LT LU LV MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK TJ TM TR TT UA UG US UZ VN
AU 9664770 A 19961230 (199716) C07K014-81
WO 9640784 A3 19970313 (199728) C07K014-81
EP 837883 A2 19980429 (199821) EN C07K014-81
R: AT BE CH DE DK ES FI FR GB GR IE IT LI LU MC NL PT SE
US 5888968 A 19990330 (199920) A61K038-36
AU 713338 B 19991202 (200008) C07K014-81
JP 11514334 W 19991207 (200008) 92 C07K014-81
AU 2000020611 A 20000615 (200036)# A61K038-57
US 6319896 B1 20011120 (200174) A61K038-16
US 6323326 B1 20011127 (200175) A23J001-00
AU 759412 B 20030417 (200333)# A61K038-57
CA 2450795 A1 19961219 (200414) EN C07K014-81
CA 2450797 A1 19961219 (200414) EN C07K014-81
CA 2450800 A1 19961219 (200414) EN A61K038-55
CA 2450804 A1 19961219 (200414) EN A61K038-55
CA 2450953 A1 19961219 (200414) EN A61K038-55
CA 2451969 A1 19961219 (200417) EN C07K014-81
CA 2451970 A1 19961219 (200417) EN C07K014-81
CA 2451973 A1 19961219 (200417) EN C07K014-81
JP 2004083591 A 20040318 (200420) 48 C07K014-81

CA 2223745 C 20040316 (200421) EN C07K014-81
AU 2003200506 A1 20030417 (200433)# A61K038-57

ADT WO 9640784 A2 WO 1996-US9980 19960607; AU 9664770 A AU 1996-64770 19960607; WO 9640784 A3 WO 1996-US9980 19960607; EP 837883 A2 EP 1996-924269 19960607, WO 1996-US9980 19960607; US 5888968 A Cont of US 1995-477677 19950607, US 1996-734997 19961022; AU 713338 B AU 1996-64770 19960607; JP 11514334 W WO 1996-US9980 19960607, JP 1997-502126 19960607; AU 2000020611 A Div ex AU 1996-64770 19960607, AU 2000-20611 20000302; US 6319896 B1 Cont of US 1995-473668 19950607, Cont of US 1995-477677 19950607, Div ex WO 1996-US9980 19960607, Div ex US 1999-973211 19990611, US 1999-443098 19991118; US 6323326 B1 CIP of US 1995-473688 19950607, CIP of US 1995-477677 19950607, WO 1996-US9980 19960607, US 1999-973211 19990611; AU 759412 B Div ex AU 1996-64770 19960607, AU 2000-20611 20000302; CA 2450795 A1 Div ex CA 1996-2223745 19960607, CA 1996-2450795 19960607; CA 2450797 A1 Div ex CA 1996-2223745 19960607, CA 1996-2450797 19960607; CA 2450800 A1 Div ex CA 1996-2223745 19960607, CA 1996-2450800 19960607; CA 2450804 A1 Div ex CA 1996-2223745 19960607, CA 1996-2450804 19960607; CA 2450953 A1 Div ex CA 1996-2223745 19960607, CA 1996-2450953 19960607; CA 2451969 A1 Div ex CA 1996-2223745 19960607, CA 1996-2451969 19960607; CA 2451970 A1 Div ex CA 1996-2223745 19960607, CA 1996-2451970 19960607; CA 2451973 A1 Div ex CA 1996-2223745 19960607, CA 1996-2451973 19960607; JP 2004083591 A Div ex JP 1997-502126 19960607, JP 2003-326585 20030918; CA 2223745 C CA 1996-2223745 19960607, WO 1996-US9980 19960607; AU 2003200506 A1 Div ex AU 2000-20611 20000302, AU 2003-200506 20030214

FDT AU 9664770 A Based on WO 9640784; EP 837883 A2 Based on WO 9640784; AU 713338 B Previous Publ. AU 9664770, Based on WO 9640784; JP 11514334 W Based on WO 9640784; AU 2000020611 A Div ex AU 713338; US 6323326 B1 CIP of US 5923306, Based on WO 9640784; AU 759412 B Previous Publ. AU 2000020611, Div ex AU 713338; CA 2223745 C Based on WO 9640784

PRAI US 1995-477677 19950607; US 1995-473668 19950607;
US 1996-734997 19961022; AU 2000-20611 20000302;
US 1999-973211 19990611; US 1999-443098 19991118;
US 1995-473688 19950607; AU 2003-200506 20030214

REP EP 131864; EP 325691; EP 473564; EP 559632

IC ICM A23J001-00; A61K038-16; A61K038-36; A61K038-55; A61K038-57; C07K014-81
ICS A01N037-18; A61K009-08; A61K031-198; A61K038-00; A61K047-04; A61K047-10; A61K047-12; A61K047-18; A61K047-20; A61K047-22; A61K047-26; A61K047-30; A61K047-34; A61K047-36; A61P007-02; A61P043-00; C07K001-00; C07K001-02; C07K001-113; C07K001-14; C07K001-16; C07K001-18; C07K001-30; C07K014-745; **C12P021-00**

AB WO 9640784 A UPAB: 20020213

An aqueous formulation comprising tissue factor pathway inhibitor (TFPI) at a concentration of > 1 mg/ml and a charged polymer (CP) is new. The charged polymer (CP) is pref. a sulphated polysaccharide (such as heparin or dextran sulphate) or a polyphosphate, pref. immobilised on a solid support. Also new are: a method of modifying the solubility of a protein having a first domain which has a net positive charge and a second domain which has a net negative charge comprises adding an aqueous solution of CP to reduce inter- and intra-molecular interactions between the charged domains; and a method of **refolding** an improperly **folded** or denatured protein (e.g. TFPI) comprises adding CP to a solution of the protein prior to allowing the protein to **refold.** Further claimed is a pharmaceutically acceptable compsn. comprising > 0.2 mg/ml TFPI and a solubilising agent chosen from acetate ions, NaCl, citrate ions, isocitrate ions, glycine, glutamate, succinate ions, histidine, imidazole and SDS.

USE - The methods are partic. useful for solubilising, formulating, purifying and **refolding** proteins (especially TFPI) which have been engineered by genetic recombination and produced in bacterial, yeast or

other cells in a form that has a non-native tertiary structure. TFPI is a coagulation inhibitor which has clot-inhibiting properties.
Dwg.0/28
FS CPI
FA AB; DCN
MC CPI: A12-V01; **B04-N03**; B04-N0300E; **D05-H13**; D05-H17

L28 ANSWER 21 OF 35 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
AN 96185290 EMBASE
DN 1996185290
TI A rapid single-step purification method for human interferon-γ from isolated Escherichia coli inclusion bodies.
AU Haelewyn J.; De Ley M.
CS Laboratorium voor Biochemie, KULeuven, Celestrijnenlaan 200 G,B-3001 Leuven-Heverlee, Belgium
SO Biochemistry and Molecular Biology International, (1995) 37/6 (1163-1171). ISSN: 1039-9712 CODEN: BMBIES
CY Australia
DT Journal; Article
FS 022 Human Genetics
029 Clinical Biochemistry
LA English
SL English
AB A fast purification method for **recombinant** human interferon-γ, produced in E.coli, was elaborated. Human IFN-γ accumulated in the cytoplasm of E.coli cells as inclusion bodies (IB). After lysis, the IB were isolated from the cell debris by means of a density gradient ultracentrifugation, and **solubilized** in 6 M guanidine hydrochloride. The subsequent refolding step was optimized for a maximal recovery of the biologically active dimer. Refolded IFN-γ was then purified to homogeneity in a single cation exchange chromatographic step, yielding 12.5 mg protein per liter E.coli culture. The dimeric nature of the refolded protein was visualized by means of interchain cross-linking. In a subsequent Western blot the resulting derivative was recognized by a panel of five monoclonal antibodies, indicating that those epitopes on the protein surface remained unaffected upon cross-linking.
CT Medical Descriptors:
*cell inclusion
*protein expression
***protein purification**
article
cytolysis
density gradient centrifugation
escherichia coli
expression vector
human
immunoblotting
ion exchange chromatography
methodology
nonhuman
protein cross linking
protein folding
Drug Descriptors:
dimer
***recombinant gamma interferon**
epitope
guanidine hydrochloride
monoclonal antibody

RN (guanidine hydrochloride) 50-01-1

L28 ANSWER 22 OF 35 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
AN 95302984 EMBASE
DN 1995302984
TI Human stress protein hsp70: Overexpression in E. coli, purification and characterization.
AU Jindal S.; Murray P.; Rosenberg S.; Young R.A.; Williams K.P.
CS PerSeptive Biosystems, 500 Old Connecticut Path,Framingham, MA, United States
SO Bio/Technology, (1995) 13/10 (1105-1109).
ISSN: 0733-222X CODEN: BTCHDA
CY United States
DT Journal; Article
FS 004 Microbiology
022 Human Genetics
029 Clinical Biochemistry
LA English
SL English
AB The gene encoding the stress-inducible member of human heat shock protein hsp70, was expressed in E. coli using the bacteriophage T7 RNA polymerase-based gene expression system. **Recombinant** hsp70 (R-hsp70) was purified from inclusion bodies after **solubilization** and refolding, using a combination of ATP-agarose affinity chromatography and ion-exchange chromatography. R-hsp70 was shown to be monomeric and free of its structurally similar E. coli counterpart, DnaK. In addition, R-hsp70 is functional as demonstrated by its ability to bind to peptides and to ATP. The availability of pure, correctly folded R-hsp70 in sufficient quantity will assist in the structural and functional characterization of hsp70. Furthermore, an understanding of the cytoprotective function of hsp70 and its role in immune responses during infections will be facilitated by the availability of pure R-hsp70.
CT Medical Descriptors:
*gene expression
*protein analysis
***protein purification**
affinity chromatography
article
cell inclusion
cell protection
escherichia coli
immune response
ion exchange chromatography
protein binding
protein folding
structure activity relation
Drug Descriptors:
*adenosine triphosphate
*heat shock protein
*rna polymerase
RN (adenosine triphosphate) 15237-44-2, 56-65-5, 987-65-5; (rna polymerase) 9014-24-8

L28 ANSWER 25 OF 35 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
AN 95001439 EMBASE
DN 1995001439
TI Approaches to analysis of aggregates and demonstrating mass balance in pharmaceutical protein (basic fibroblast growth factor)

formulations.
AU Shahrokh Z.; Stratton P.R.; Eberlein G.A.; Wang Y.J.
CS Department of Pharmaceutical R and D, Genetech Inc., 460 Point San Bruno Blvd.,South San Francisco, CA, United States
SO Journal of Pharmaceutical Sciences, (1994) 83/12 (1645-1650).
ISSN: 0022-3549 CODEN: JPMSAE
CY United States
DT Journal; Conference Article
FS 029 Clinical Biochemistry
030 Pharmacology
037 Drug Literature Index
LA English
SL English
AB Denaturation, aggregation, and precipitation, which are common events in protein aging, limit the development of pharmaceutical protein **formulations.** Using the example of basic fibroblast growth factor (bFGF), we describe a systematic approach for quantitative recovery of soluble and insoluble aggregates in aged samples to achieve mass balance in three analytical methods. UV spectroscopy, size exclusion HPLC (HP-SEC), and reverse phase HPLC. Soluble aggregates were evaluated by UV spectroscopy and HP-SEC; the latter method was modified to include 2 M guanidine hydrochloride (GnHCl) in the mobile phase in order to differentiate and simultaneously analyze native and denatured protein. Insoluble aggregates were first **solubilized** with GnHCl and then recovered quantitatively with the modified HP-SEC method. Chaotrope treatment did not affect the UV peak absorbance but altered the HPLC profiles. The changes were consistent with dissociation of disulfide-linked multimers to monomers with an intramolecular disulfide linkage. This phenomenon was used for estimation of the monomer-multimer distribution in the untreated aggregated sample. These methods established approaches for quantitative recovery and characterization of bFGF aggregates.
CT Medical Descriptors:
***drug formulation**
***protein purification**
conference paper
controlled study
high performance liquid chromatography
mass
precipitation
protein analysis
protein denaturation
protein folding
quantitative assay
solubility
spectroscopy
structure analysis
Drug Descriptors:
***recombinant basic fibroblast growth factor: AN, drug analysis**
***recombinant basic fibroblast growth factor: PR, pharmaceutics**
guanidine hydrochloride
RN (guanidine hydrochloride) 50-01-1

L28 ANSWER 29 OF 35 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
AN 93239715 EMBASE
DN 1993239715
TI Production and characterization of an analog of acidic fibroblast growth factor with enhanced stability and biological activity.
AU Arakawa T.; Horan T.P.; Narhi L.O.; Rees D.C.; Schiffer S.G.; Holst P.L.;

Prestrelski S.J.; Tsai L.B.; Fox G.M.
CS Amgen Inc, Amgen Center,Thousand Oaks, CA 91320-1789, United States
SO Protein Engineering, (1993) 6/5 (541-546).
ISSN: 0269-2139 CODEN: PRENE
CY United Kingdom
DT Journal; Article
FS 022 Human Genetics
029 Clinical Biochemistry
LA English
SL English
AB We have used **recombinant** DNA methods to produce two forms of bovine acidic fibroblast growth factor (aFGF), one with alanine substituted for the cysteine at position 47 and the other with the Ala47 change plus the substitution of glycine for the naturally occurring histidine at position 93. Both forms were expressed at high levels in Escherichia coli and purified to near homogeneity by **solubilization** of the inclusion bodies containing the aFGF, ion exchange chromatography, refolding of the protein and hydrophobic interaction chromatography. Circular dichroic and infrared spectra suggested that the proteins are similar in secondary and tertiary structures and contain little or no α-helical conformations. Hydrophobic interaction chromatography showed that aFGF C47A/H93G is slightly more hydrophobic than the aFGF C47A form, suggesting that residue 93 is exposed to the solvent. Half-maximal activity in an in vitro bioassay system was reached at a 10- to 20-fold lower dose for the aFGF C47A/H93G form than for the aFGF C47A form, suggesting that alteration of this residue has an effect on the region responsible for receptor binding. Addition of 50 μg/ml heparin enhanced the in vitro activity of the aFGFs, reducing the half-maximal dose to approximately 100 pg/ml for both forms, comparable to that observed previously for basic FGF with or without heparin in this assay system.
CT Medical Descriptors:
*protein stability
amino acid substitution
article
circular dichroism
cow
escherichia coli
gene expression
hydrophobicity
infrared spectrometry
nonhuman
protein folding
protein purification
protein secondary structure
protein structure
Drug Descriptors:
*acidic fibroblast growth factor
alanine
cysteine
glycine
heparin
histidine
protein derivative
recombinant dna
RN (acidic fibroblast growth factor) 106096-92-8; (alanine) 56-41-7, 6898-94-8; (cysteine) 4371-52-2, 52-89-1, 52-90-4; (glycine) 56-40-6, 6000-43-7, 6000-44-8; (heparin) 37187-54-5, 8057-48-5, 8065-01-8, 9005-48-5; (histidine) 645-35-2, 7006-35-1, 71-00-1

L28 ANSWER 33 OF 35 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED.
on STN
AN 92141536 EMBASE
DN 1992141536
TI **Solubilization** and regeneration of Vitreoscilla hemoglobin isolated from protein inclusion bodies.
AU Hart R.A.; Bailey J.E.
CS Div. Chemistry/Chemical Engineering, California Institute of Technology,Pasadena, CA 91125, United States
SO Biotechnology and Bioengineering, (1992) 39/11 (1112-1120).
ISSN: 0006-3592 CODEN: BIBIAU
CY United States
DT Journal; Article
FS 004 Microbiology
029 Clinical Biochemistry
LA English
SL English
AB Vitreoscilla hemoglobin (VHb), a homodimeric protein containing two heme groups in its native state, was used as a model to investigate inclusion body apoprotein **solubilization**, prosthetic group incorporation, and reactivation. High-level expression in **recombinant** Escherichia coli results in accumulation of a substantial portion of heme-free VHb in inclusion bodies. VHb can be **solubilized** from these inclusion bodies by relatively low concentrations of urea with the dissolution midpoint at approximately 3.2M urea. Dissolution in the presence of stoichiometric heme shifts the dissolution midpoint to approximately 4.5M urea without influencing the dissolution properties of contaminant proteins, suggesting the effect is specific for VHb. Denaturation of apoVHb and holoVHb obtained from purified native VHb has midpoints of 2.9M and 5.1M urea, respectively. VHb **solubilized** from inclusion bodies with urea at concentrations from 0 to 3.5M urea can be regenerated by heme addition without dilution of urea to yield active holoVHb. The fraction of **solubilized** VHb reconstituted upon heme addition is maximum at around 30% when **solubilization** and reconstitution is conducted in less than 1M urea. At these low urea concentrations, approximately 5% of inclusion body VHb is **solubilized.** These results show the utility of prosthetic group addition to reconstitute holoVHb in the presence of urea. Also, these findings suggest that some inclusion body protein has partially folded conformation and that a fractional dissolution and refolding process may be advantageous.
CT Medical Descriptors:
*cell inclusion
*protein isolation
animal tissue
article
escherichia coli
gene expression
nonhuman
protein folding
protein purification
Drug Descriptors:
*hemoglobin: EC, endogenous compound
urea
RN (hemoglobin) 9008-02-0; (urea) 57-13-6

L28 ANSWER 34 OF 35 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED.
on STN
AN 92226283 EMBASE
DN 1992226283

TI Expression and purification of biologically active human OSF-1 in Escherichia coli.
AU Takamatsu H.; Itoh M.; Kimura M.; Gospodarowicz D.; Amann E.
CS Hoechst Japan Limited, Pharma Research Laboratories, 3-2, Minamidai 1-chome,Kawagoe-City, Saitama 350, Japan
SO Biochemical and Biophysical Research Communications, (1992) 185/1 (224-230).
ISSN: 0006-291X CODEN: BBRCA
CY United States
DT Journal; Article
FS 029 Clinical Biochemistry
LA English
SL English
AB OSF-1 (also known as pleiotrophin, HB-GAM, HBGF-8 or HBNF) is a heparin-binding, neurotrophic protein. Its tissue-specific expression in rats is developmentally regulated and the protein is highly conserved between species. The protein is believed to be involved in neuronal development. Previous experiments in our laboratory showed that OSF-1 is primarily expressed in brain and bone. The biological function of OSF-1 in bone is unknown. In order to overcome the limited availability of the native protein, we now report on the high-level expression of human OSF-1 in Escherichia coli. The protein is present in the form of inclusion bodies, which were isolated and **solubilized.** The partially purified protein was refolded and further purified employing heparin sepharose chromatography. N-terminal sequence determination revealed the same amino acid sequence as the natural mature protein. The isolated backfolded **recombinant** human OSF-1 did promote neurites outgrowth in primary cultures of cortical neurons.
CT Medical Descriptors:
*gene expression
*protein analysis
***protein purification**
amino terminal sequence
article
bone
brain
escherichia coli
nerve cell differentiation
neurite
priority journal
protein folding
tissue specificity
Drug Descriptors:
*heparin binding protein
recombinant protein

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PLEASE SEE "HELP USAGETERMS" FOR DETAILS.
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FILE COVERS 1907 - 9 Jul 2004  VOL 141 ISS 3
FILE LAST UPDATED: 8 Jul 2004  (20040708/ED)

 This file contains CAS Registry Numbers for easy and accurate
 substance identification.

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                   /BI
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L16           18 SEA FILE=HCAPLUS ABB=ON  PLU=ON  L15 AND (PY<=1995 OR PRY<=1995
                   OR AY<=1995)
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                   TERTIARY/OBI)


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FROM JANUARY 1969 TO DATE.

RECORDS LAST ADDED: 7 July 2004 (20040707/ED)

FILE RELOADED: 19 October 2003.

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L39          28 SEA FILE=BIOSIS ABB=ON  PLU=ON  METHOD? AND L38
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                  OR "TISSUE FACTOR PATHWAY INHIBITOR-2 (HUMAN)"/CN
L29 (       539)SEA FILE=BIOSIS ABB=ON  PLU=ON  L28
L30 (        46)SEA FILE=BIOSIS ABB=ON  PLU=ON  TISSUE FACTOR INHIBITOR?
L31 (       571)SEA FILE=BIOSIS ABB=ON  PLU=ON  L29 OR L30
L32 (       125)SEA FILE=BIOSIS ABB=ON  PLU=ON  (BLOOD(W)COAGULATION)(3A)(EPI
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L36 (    483873)SEA FILE=BIOSIS ABB=ON  PLU=ON  ?PURIF? OR ?SOLUBIL? OR
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L37 (      1191)SEA FILE=BIOSIS ABB=ON  PLU=ON  TFPI OR L35
L38 (        80)SEA FILE=BIOSIS ABB=ON  PLU=ON  L36 AND L37
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L49        5662 SEA FILE=BIOSIS ABB=ON  PLU=ON  ARGININE/CT
L51           0 SEA FILE=BIOSIS ABB=ON  PLU=ON  L49 AND L46


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L59           2 SEA FILE=EMBASE ABB=ON  PLU=ON  L56 AND L58
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L61 (    246440)SEA FILE=EMBASE ABB=ON  PLU=ON  (RECOMB? OR ENGINEER?)
L62 (      5086)SEA FILE=EMBASE ABB=ON  PLU=ON  L61 AND L60
L63 (     20401)SEA FILE=EMBASE ABB=ON  PLU=ON  "PROTEIN FOLDING"/CT
L64 (       305)SEA FILE=EMBASE ABB=ON  PLU=ON  L62 AND L63
L65 (        58)SEA FILE=EMBASE ABB=ON  PLU=ON  L64 AND PY<=1995
L66 (    159116)SEA FILE=EMBASE ABB=ON  PLU=ON  ?SOLUBIL? OR ?FORMULA?
L67          13 SEA FILE=EMBASE ABB=ON  PLU=ON  L66 AND L65
L68          15 SEA FILE=EMBASE ABB=ON  PLU=ON  L59 OR L67
L69       22826 SEA FILE=EMBASE ABB=ON  PLU=ON  ARGININE/CT,CW
L70           0 SEA FILE=EMBASE ABB=ON  PLU=ON  L68 AND L69


=> d que l72
L52 (       689)SEA FILE=EMBASE ABB=ON  PLU=ON  ANTITHROMBOPLASTIN/CT OR
                "EXTRINSIC COAGULATION PATHWAY INHIBITOR"/CT OR "LIPOPROTEIN
                ASSOCIATED COAGULATION INHIBITOR"/CT OR TFPI
L53 (      6435)SEA FILE=EMBASE ABB=ON  PLU=ON  PURIFICATION/CT
L54 (     24210)SEA FILE=EMBASE ABB=ON  PLU=ON  "PROTEIN PURIFICATION"/CT
L55 (     30595)SEA FILE=EMBASE ABB=ON  PLU=ON  L53 OR L54
L56 (        13)SEA FILE=EMBASE ABB=ON  PLU=ON  L52 AND L55
L57 (      3653)SEA FILE=EMBASE ABB=ON  PLU=ON  SOLUBILIZATION/CT
L58 (     55767)SEA FILE=EMBASE ABB=ON  PLU=ON  ?SOLUBIL? OR L57
L59           2 SEA FILE=EMBASE ABB=ON  PLU=ON  L56 AND L58
L60 (     24210)SEA FILE=EMBASE ABB=ON  PLU=ON  "PROTEIN PURIFICATION"/CT
L61 (    246440)SEA FILE=EMBASE ABB=ON  PLU=ON  (RECOMB? OR ENGINEER?)
L62 (      5086)SEA FILE=EMBASE ABB=ON  PLU=ON  L61 AND L60
L63 (     20401)SEA FILE=EMBASE ABB=ON  PLU=ON  "PROTEIN FOLDING"/CT
L64 (       305)SEA FILE=EMBASE ABB=ON  PLU=ON  L62 AND L63
L65 (        58)SEA FILE=EMBASE ABB=ON  PLU=ON  L64 AND PY<=1995
L66 (    159116)SEA FILE=EMBASE ABB=ON  PLU=ON  ?SOLUBIL? OR ?FORMULA?
L67          13 SEA FILE=EMBASE ABB=ON  PLU=ON  L66 AND L65
L68          15 SEA FILE=EMBASE ABB=ON  PLU=ON  L59 OR L67
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L71         6231 SEA FILE=EMBASE ABB=ON  PLU=ON  TERTIARY(A)STRUCTURE
L72            1 SEA FILE=EMBASE ABB=ON  PLU=ON  L68 AND L71


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FILE LAST UPDATED:          2 JUL 2004      <20040702/UP>
MOST RECENT DERWENT UPDATE:     200442        <200442/DW>
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    NEW FORMAT GERMAN PATENT APPLICATION AND PUBLICATION
    NUMBERS. SEE ALSO:
    http://www.stn-international.de/archive/stnews/news0104.pdf <<<

=> d que 191
L73 (        87) SEA FILE=WPIX ABB=ON  PLU=ON  TISSUE(W)FACTOR(W)PATHWAY(W)INHIB
                 ITOR/BIX
L74 (         7) SEA FILE=WPIX ABB=ON  PLU=ON  (BLOOD(W)COAGULATION(3A)(EPI OR
                 LACI OR TFI OR EXTRINSIC OR LIPOPROTEIN?))/BIX
L75 (        94) SEA FILE=WPIX ABB=ON  PLU=ON  L73 OR L74
L76 (    108459) SEA FILE=WPIX ABB=ON  PLU=ON  PURIFICATION/BIX
L77 (    876551) SEA FILE=WPIX ABB=ON  PLU=ON  (?SOLUB? OR ?FORMULA?)/BIX
L78 (    176967) SEA FILE=WPIX ABB=ON  PLU=ON  PURIF?/BIX
L79 (    176967) SEA FILE=WPIX ABB=ON  PLU=ON  L76 OR L78
L80 (   1015415) SEA FILE=WPIX ABB=ON  PLU=ON  L79 OR L77
L81          35 SEA FILE=WPIX ABB=ON  PLU=ON  L80 AND L75
L82 (      4315) SEA FILE=WPIX ABB=ON  PLU=ON  (B04-B04A5 OR B04-N03)/MC
L83 (     12468) SEA FILE=WPIX ABB=ON  PLU=ON  D05-H13/MC
L84 (       267) SEA FILE=WPIX ABB=ON  PLU=ON  L82 AND L83
L85 (      7491) SEA FILE=WPIX ABB=ON  PLU=ON  C12P021-00/IC
L86 (        47) SEA FILE=WPIX ABB=ON  PLU=ON  L84 AND L85
L87 (    238921) SEA FILE=WPIX ABB=ON  PLU=ON  ?FOLD?/BIX
L88           3 SEA FILE=WPIX ABB=ON  PLU=ON  L86 AND L87
L89          37 SEA FILE=WPIX ABB=ON  PLU=ON  L81 OR L88
L90         389 SEA FILE=WPIX ABB=ON  PLU=ON  TERTIARY(2A)STRUCTURE/BIX
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L91            1 SEA FILE=WPIX ABB=ON  PLU=ON  L89 AND L90


=> d que l93
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                 ITOR/BIX
L74 (          7)SEA FILE=WPIX ABB=ON  PLU=ON  (BLOOD(W)COAGULATION(3A)(EPI OR
                 LACI OR TFI OR EXTRINSIC OR LIPOPROTEIN?))/BIX
L75 (         94)SEA FILE=WPIX ABB=ON  PLU=ON  L73 OR L74
L76 (     108459)SEA FILE=WPIX ABB=ON  PLU=ON  PURIFICATION/BIX
L77 (     876551)SEA FILE=WPIX ABB=ON  PLU=ON  (?SOLUB? OR ?FORMULA?)/BIX
L78 (     176967)SEA FILE=WPIX ABB=ON  PLU=ON  PURIF?/BIX
L79 (     176967)SEA FILE=WPIX ABB=ON  PLU=ON  L76 OR L78
L80 (    1015415)SEA FILE=WPIX ABB=ON  PLU=ON  L79 OR L77
L81            35 SEA FILE=WPIX ABB=ON  PLU=ON  L80 AND L75
L82 (       4315)SEA FILE=WPIX ABB=ON  PLU=ON  (B04-B04A5 OR B04-N03)/MC
L83 (      12468)SEA FILE=WPIX ABB=ON  PLU=ON  D05-H13/MC
L84 (        267)SEA FILE=WPIX ABB=ON  PLU=ON  L82 AND L83
L85 (       7491)SEA FILE=WPIX ABB=ON  PLU=ON  C12P021-00/IC
L86 (         47)SEA FILE=WPIX ABB=ON  PLU=ON  L84 AND L85
L87 (     238921)SEA FILE=WPIX ABB=ON  PLU=ON  ?FOLD?/BIX
L88            3 SEA FILE=WPIX ABB=ON  PLU=ON  L86 AND L87
L89           37 SEA FILE=WPIX ABB=ON  PLU=ON  L81 OR L88
L92         6718 SEA FILE=WPIX ABB=ON  PLU=ON  ARGININE/BIX
L93            5 SEA FILE=WPIX ABB=ON  PLU=ON  L89 AND L92


=> dup rem l72 l27 l91 l93
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PROCESSING COMPLETED FOR L72
PROCESSING COMPLETED FOR L27
PROCESSING COMPLETED FOR L91
PROCESSING COMPLETED FOR L93
L94            9 DUP REM L72 L27 L91 L93 (0 DUPLICATES REMOVED)

=> d all l94 1-9

L94  ANSWER 1 OF 9  WPIX COPYRIGHT 2004 THOMSON DERWENT on STN
AN   2003-902745 [82]    WPIX
DNC  C2003-256253
TI   Composition useful for the treatment of pathological condition e.g.
     restenosis, diabetic retinopathy and cancer comprises a protein, at least
     one linker and a cytotoxic compound.
DC   B03 B04 B05
IN   LIOTTA, D; SHOJI, M; SNYDER, J; SUN, A; LIOTTA, D C; SYNDER, J
PA   (LIOT-I) LIOTTA D; (SHOJ-I) SHOJI M; (SNYD-I) SNYDER J; (SUNA-I) SUN A;
     (UYEM-N) UNIV EMORY
CYC  103
PI   WO 2003075847   A2 20030918 (200382)* EN   81    A61K000-00
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            LU MC MW MZ NL OA PT RO SD SE SI SK SL SZ TR TZ UG ZM ZW
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W: AE AG AL AM AT AU AZ BA BB BG BR BY BZ CA CH CN CO CR CU CZ DE DK DM DZ EC EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX MZ NI NO NZ OM PH PL PT RO RU SC SD SE SG SK SL TJ TM TN TR TT TZ UA UG UZ VC VN YU ZA ZM ZW

US 2004009914 A1 20040115 (200406) A61K038-17
AU 2003220091 A1 20030922 (200431) A61K000-00

ADT WO 2003075847 A2 WO 2003-US7043 20030307; US 2004009914 A1 Provisional US 2002-362762P 20020308, US 2003-383898 20030307; AU 2003220091 A1 AU 2003-220091 20030307

FDT AU 2003220091 A1 Based on WO 2003075847

PRAI US 2002-362762P 20020308; US 2003-383898 20030307

IC ICM A61K000-00; A61K038-17

AB WO2003075847 A UPAB: 20031223

NOVELTY - A composition (C) comprises a protein selectively binding a surface marker of a target cell, at least one linker covalently bonded to the protein and a cytotoxic compound bonded to the linker by a hydrolyzable bond.

DETAILED DESCRIPTION - An INDEPENDENT CLAIM is included for the production of a cytotoxic compound-protein conjugate (A) involves synthesizing a product (P) comprising a cytotoxic compound, bonding covalently (P) and the linker followed by covalently bonding at least one molecule (M) of the composition to a protein.

ACTIVITY - Vasotropic; Antidiabetic; Ophthalmological; Antirheumatic; Antiarthritic; Dermatological; Antiinflammatory; Cytostatic.

3,5-Bis-(2-fluoro-benzylidene)-piperidin-4-one (EF24) was 10 times more effective than either cisplatin or curcumin when tested against tumor cells in the NCI screening system.

MECHANISM OF ACTION - Pathological condition modulator; Angiogenesis inhibitor; Vascular endothelial growth factor receptor antagonist.

USE - For modulating a pathological condition and proliferation of the cell in the treatment of e.g. hypercoagulapathy, restenosis, diabetic retinopathy, rheumatoid arthritis, skin disorder inflammation and cancer (e.g. leukemia, breast cancer, lung cancer, liver cancer, melanoma and prostrate cancer) (claimed).

ADVANTAGE - The composition is antiangiogenic and reduces the proliferation of a target cell thus causing reduction in a tumor; increases the efficacy of the cytotoxic agent and decreases the side effects by delivering the agent to target cells by binding the composition to the surface maker on the target cells as the composition is internalized by the target cell. Curcumin analog (preferably EF24) was 10 times more potent than cisplatin which is well-known anticancer agent and the conjugates kill cancer cells and vascular endothelial cells that express tissue factor.

Dwg.0/5

FS CPI

FA AB; GI; DCN

MC CPI: B07-A02B; B07-A03; B07-B02; B07-D05; B07-D06; B10-A15; B10-A16; B10-A17; B14-C03; B14-C09B; B14-F02; B14-F04; B14-H01; B14-H01A; B14-N03; B14-N17; B14-S04

L94 ANSWER 2 OF 9 WPIX COPYRIGHT 2004 THOMSON DERWENT on STN

AN 2003-902653 [82] WPIX

CR 2003-689387 [65]; 2003-897354 [82]; 2003-897355 [82]

DNC C2003-256173

TI Treating sepsis involves continuous intravenous infusion of **tissue factor pathway inhibitor** or **tissue factor pathway inhibitor** analog to a patient.

DC B04 D16

IN CREASEY, A A

PA (CHIR) CHIRON CORP
CYC 101
PI WO 2003055442 A2 20030710 (200382)* EN 58 A61K000-00
RW: AT BE BG CH CY CZ DE DK EA EE ES FI FR GB GH GM GR IE IT KE LS LU MC MW MZ NL OA PT SD SE SK SL SZ TR TZ UG ZM ZW
W: AE AG AL AM AT AU AZ BA BB BG BR BY BZ CA CH CN CO CR CU CZ DE DK DM DZ EC EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX MZ NO NZ OM PH PL PT RO RU SD SE SG SI SK SL TJ TM TN TR TT TZ UA UG US UZ VC VN YU ZA ZM ZW
AU 2002365131 A1 20030715 (200421) A61K000-00
ADT WO 2003055442 A2 WO 2002-US32625 20021015; AU 2002365131 A1 AU 2002-365131 20021015
FDT AU 2002365131 A1 Based on WO 2003055442
PRAI US 2001-328806P 20011015
IC ICM A61K000-00
AB WO2003055442 A UPAB: 20040326
NOVELTY - Treating (M1) sepsis comprising continuous intravenous infusion of **tissue factor pathway inhibitor** (TFPI) or TFPI analog to a patient at a dose rate equivalent to administration of reference ala-TFPI, which is from 0.00025 to 0.050 mg/kg/hr for a period of at least 72 hours, is new.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for:

(1) decreasing (M2) the risk and severity of sepsis comprising continuous intravenous infusion of TFPI or TFPI analog to a patient at a dose rate equivalent to administration of reference ala-TFPI, which is from 0.00025 to 0.050 mg/kg/hr for a period of at least 72 hours;

(2) treating (M3) acute inflammation, including sepsis and septic shock, comprising continuous intravenous infusion of TFPI or a TFPI analog to a patient at a dose rate equivalent to administration of reference ala-TFPI, which is from 0.00025 to 0.050 mg/kg/hour and an additional agent chosen from an antibiotic, a monoclonal antibody, a cytokine inhibitor, and a complement inhibitor; and

(3) treating (M4) a disease state not associated with disseminated intravascular coagulation (DIC) and in which TNF, interleukin (IL)-1, or another cytokine up-regulates tissue factor, comprising continuous intravenous infusion of an agent chosen from TFPI or a TFPI analog to a patient at a dose rate equivalent to administration of reference ala-TFPI, which is from 0.00025 to 0.050 mg/kg/hr for 72 hours.

ACTIVITY - Antibacterial; Immunosuppressive; Antiinflammatory; Respirator-Gen..

To detect the effectiveness of low dose of TFPI in rabbit peritonitis model was as follows. Treatment with gentamicin was initiated 4 hour after induction of peritonitis. After 4 hour, clinical symptoms of fever, chills, and a slight drop in blood pressure typically were present. Rabbits were randomized to receive a 24-hour infusion of placebo or one of six doses of ala-TFPI, consisting of a bolus dose followed by continuous intravenous infusion. Rabbits remaining alive after seven days were considered survivors. The cause of death was respiratory and multiple organ failure. At necropsy, the lungs of non survivors were grossly edematous, and the airways were filled with pink, frothy fluid. The kidneys were hemorrhagic and congested. The liver typically contained abscesses that grew out Escherichia coli. Abscesses were present throughout the peritoneal cavity. In contrast, 7-day survivors showed evidence of minimal pulmonary congestion. Small scattered abscesses often were present in the peritoneal cavity, but other solid organs (adrenals, liver, and spleen) were grossly normal.

MECHANISM OF ACTION - Inhibitor of acute or chronic inflammation.

USE - (M1) is useful for treating a patient suffering from sepsis, shock, or acute respiratory distress syndrome (ARDS). (M3) is useful for

treating acute inflammation. (M4) is useful for treating chronic or acute inflammation.
Dwg.1/3
FS CPI
FA AB; GI; DCN
MC CPI: B04-G01; B04-H01; B04-N06; B14-C03; B14-K01F; B14-L06; B14-L07; B14-S06; D05-H11

L94 ANSWER 3 OF 9 WPIX COPYRIGHT 2004 THOMSON DERWENT on STN
AN 2003-689387 [65] WPIX
CR 2003-897354 [82]; 2003-897355 [82]; 2003-902653 [82]
DNC C2003-188883
TI Treating or preventing severe pneumonia comprises administering **tissue factor pathway inhibitor** or an analog of it, and preferably an additional agent, such as a monoclonal antibody.
DC B04 D16
IN CREASEY, A A
PA (CHIR) CHIRON CORP
CYC 101
PI WO 2003032904 A2 20030424 (200365)* EN 39 A61K000-00
RW: AT BE BG CH CY CZ DE DK EA EE ES FI FR GB GH GM GR IE IT KE LS LU MC MW MZ NL OA PT SD SE SK SL SZ TR TZ UG ZM ZW
W: AE AG AL AM AT AU AZ BA BB BG BR BY BZ CA CH CN CO CR CU CZ DE DK DM DZ EC EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX MZ NO NZ OM PH PL PT RO RU SD SE SG SI SK SL TJ TM TN TR TT TZ UA UG US UZ VC VN YU ZA ZM ZW
ADT WO 2003032904 A2 WO 2002-US32624 20021015
PRAI US 2001-328806P 20011015
IC ICM A61K000-00
AB WO2003032904 A UPAB: 20031223
NOVELTY - Treating or preventing severe pneumonia comprises administering **tissue factor pathway inhibitor** (TFPI) or a TFPI analog to a patient who has or who is at risk of having severe pneumonia.
ACTIVITY - Antiinflammatory. No suitable biological data is given.
MECHANISM OF ACTION - **Tissue factor pathway inhibitor.**
USE - The method is used for treating or preventing severe pneumonia (claimed).
ADVANTAGE - The TFPI or analog is administered at a low dose which is efficient at treating severe pneumonia and adverse side effects, such as bleeding, are minimized.
Dwg.0/0
FS CPI
FA AB; DCN
MC CPI: B04-G02; B04-G21; B04-H01; B04-H0100E; B14-F04; B14-K01; B14-L06; D05-C12; D05-H11A; D05-H17A2

L94 ANSWER 4 OF 9 WPIX COPYRIGHT 2004 THOMSON DERWENT on STN
AN 2003-897354 [82] WPIX
CR 2003-689387 [65]; 2003-897355 [82]; 2003-902653 [82]
DNC C2003-254735
TI Treating or preventing severe pneumonia by administering **tissue factor pathway inhibitor** (TFPI) or its analog to a patient having or is at risk of having severe pneumonia.
DC B04
IN CREASEY, A
PA (CREA-I) CREASEY A

CYC 1
PI US 2003139339 A1 20030724 (200382)* 14 A61K038-17
ADT US 2003139339 A1 Provisional US 2001-328806P 20011015, US 2002-270478 20021015
PRAI US 2001-328806P 20011015; US 2002-270478 20021015
IC ICM A61K038-17
AB US2003139339 A UPAB: 20031223
NOVELTY - Treating or preventing severe pneumonia comprises administering **tissue factor pathway inhibitor** (TFPI) or its analog to a patient having or is at risk of having severe pneumonia.
ACTIVITY - Antiinflammatory.
MECHANISM OF ACTION - Gene therapy; Vaccine.
USE - The method is useful for treating or preventing severe pneumonia (claimed).
Dwg.0/0
FS CPI
FA AB; DCN
MC CPI: B04-C02E1; B04-G01; B04-G21; B04-N04; B14-D03; B14-L06; B14-L07; B14-S03; B14-S11

L94 ANSWER 5 OF 9 WPIX COPYRIGHT 2004 THOMSON DERWENT on STN
AN 2001-308197 [32] WPIX
DNC C2001-095201
TI New combination of an amino acid base buffered by an acid free of its salt form increases the stability of polypeptides in pharmaceutical compositions whilst increasing isotonicity to reduce pain during administration.
DC B04
IN CHEN, B; HORA, M; HORA, M S
PA (CHIR) CHIRON CORP
CYC 95
PI WO 2001024814 A1 20010412 (200132)* EN 84 A61K038-20
RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW MZ NL OA PT SD SE SL SZ TZ UG ZW
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AU 2000078475 A 20010510 (200143) A61K038-20
NO 2002001567 A 20020522 (200247) A61K000-00
EP 1220682 A1 20020710 (200253) EN A61K038-20
R: AL AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU LV MC MK NL PT RO SE SI
BR 2000014486 A 20020917 (200264) A61K038-20
CZ 2002001186 A3 20021113 (200282) A61K038-20
HU 2002003133 A2 20021228 (200308) A61K038-20
JP 2003510368 W 20030318 (200321) 91 A61K038-00
US 6525102 B1 20030225 (200323) A01N025-00
CN 1402640 A 20030312 (200339) A61K038-20
US 2003180253 A1 20030925 (200364) A61K038-20
ADT WO 2001024814 A1 WO 2000-US27156 20001003; AU 2000078475 A AU 2000-78475 20001003; NO 2002001567 A WO 2000-US27156 20001003, NO 2002-1567 20020403; EP 1220682 A1 EP 2000-968584 20001003, WO 2000-US27156 20001003; BR 2000014486 A BR 2000-14486 20001003, WO 2000-US27156 20001003; CZ 2002001186 A3 WO 2000-US27156 20001003, CZ 2002-1186 20001003; HU 2002003133 A2 WO 2000-US27156 20001003, HU 2002-3133 20001003; JP 2003510368 W WO 2000-US27156 20001003, JP 2001-527813 20001003; US 6525102 B1 Provisional US 1999-157696P 19991004, US 2000-677643 20001003; CN 1402640 A CN 2000-816328 20001003; US 2003180253 A1 Provisional US

1999-157696P 19991004, Cont of US 2000-677643 20001003, US 2002-299039 20021118

FDT AU 2000078475 A Based on WO 2001024814; EP 1220682 A1 Based on WO 2001024814; BR 2000014486 A Based on WO 2001024814; CZ 2002001186 A3 Based on WO 2001024814; HU 2002003133 A2 Based on WO 2001024814; JP 2003510368 W Based on WO 2001024814; US 2003180253 A1 Cont of US 6525102

PRAI US 1999-157696P 19991004; US 2000-677643 20001003; US 2002-299039 20021118

IC ICM A01N025-00; A61K000-00; A61K038-00; A61K038-20
ICS A61K009-08; A61K009-20; A61K038-17; A61K038-21; A61K038-57; A61K039-00; A61K045-00; A61K047-04; A61K047-12; A61K047-18; A61P035-00; A61P043-00; C07K001-02; C07K017-00

AB WO 200124814 A UPAB: 20021031
NOVELTY - Stabilized liquid pharmaceutical composition (I) comprising a polypeptide or its variant, an amino acid base comprising **arginine**, lysine, aspartic acid or glutamic acid and a buffering agent.

DETAILED DESCRIPTION - New stabilized liquid pharmaceutical composition (I) comprises:

(a) an active agent consisting polypeptide (or one of its variants), exhibiting aggregate formation during storage in a liquid **formulation**;

(b) an amino acid base present in an amount sufficient to decrease aggregate formation of the polypeptide or its variant during storage of the composition, the amino acid base comprising at least one amino acid selected from **arginine**, lysine, aspartic acid and glutamic acid; and

(c) a buffering agent selected from an acid free of its salt forms and/or an acid in its salt form.

INDEPENDENT CLAIMS are also included for:

(1) a stabilized liquid pharmaceutical composition comprising interleukin-2 (IL-2) or one of its variants, **arginine** in its free base form (150-350 mM) and succinic acid substantially free of its salt form (80-190 mM);

(2) a stabilized liquid pharmaceutical composition comprising **tissue factor pathway inhibitor** (TFPI) or one of its variants, **arginine** in its free base form (175-325 mM) and succinic acid substantially free of its salt form (80-190 mM);

(3) a stabilized liquid pharmaceutical composition comprising **tissue factor pathway inhibitor** (TFPI) or one of its variants, **arginine** in its free base form (175-400 mM) and citric acid substantially free of its salt form (40-200 mM); and

(4) a method for increasing the stability of a polypeptide or one of its variants in a liquid pharmaceutical composition, where the polypeptide or it variant exhibits aggregate formation during storage in a liquid **formulation**, by incorporating into the composition an amino acid base in an amount sufficient to decrease aggregate formation of the polypeptide or one of its variants and a buffer selected from acids free of their salt forms, acids in their salt forms and a mixture of an acids and its salt form, the amino acid base comprising at least one amino acid selected form **arginine**, lysine, aspartic acid and glutamic acid.

USE - The invention is for stabilizing polypeptides such as IL-2 (used in the treatment of cancer metastasis) in pharmaceutical compositions.

ADVANTAGE - The increased storage stability of the composition is achieved through the influence of the amino acid on the stability of the therapeutically active polypeptide, in particular through its influence on polypeptide aggregation during storage in liquid **formulations.** The incorporation of an amino acid base and an acid free of its salt form

within liquid polypeptide-containing **formulations** results in liquid pharmaceutical compositions that are near isotonic without having to include additional isotonizing agents. Isotonicity reduces pain upon administration and the compositions of the invention exhibit reduced pain associated with burning and stinging relative to injection of normal saline. The novel combination of the invention allows for **formulations** with higher concentrations of the stabilizing amino acid than can be achieved with the use of a buffer system that is a mixture of an acid and its salt form. The higher concentration of the stabilizing amino acid allows for even greater increases in polypeptide stability, and thus increased storage stability of the **formulation**. In a stability study, **tissue factor pathway inhibitor** was **formulated** in 0.15 mg/ml final concentration in various **formulations** containing either L-**arginine** base or L-**arginine** hydrochloride. L-**arginine** hydrochloride **formulations** were buffered to pH 5.5 by citric acid or succinic acid (10 mM) in combination with its respective conjugate salt. L-**arginine** base **formulation** was titrated to pH 5.5 by either citric or succinic acid. It was found that acid titration with either succinic or citric acid allowed for a greater concentration of **arginine** in the **formulation** while maintaining isotonicity. However one of these **formulations** using 10 mM citric acid and sodium citrate to buffer 300 mM L-**arginine** hydrochloride to pH 5.5. had a solution osmolarity of 497 mOsm/kg which constitutes a hypertonic **formulation** which is not preferable for injection. On the other hand, a **formulation** using 121 mM citric acid in combination with 300 mM L-**arginine** base to adjust the pH to 5.5 had a solution osmolarity of 295 mOsm/kg, which is very close to an isotonic solution and preferable for injection. Dwg.0/11

FS CPI
FA AB; DCN
MC CPI: B04-C01; B04-H02B; B04-H02B0E; B04-N04; B04-N0400E; B10-A17; B10-A18; B10-B02J; B10-C02; B14-L06

L94 ANSWER 6 OF 9 HCAPLUS COPYRIGHT 2004 ACS on STN
AN 1999:459706 HCAPLUS
DN 131:262539
ED Entered STN: 28 Jul 1999
TI **Solubility** of Recombinant Human Tissue Factor Pathway Inhibitor
AU Chen, Bao-Lu; Wu, Xiaorong; Babuka, Susan J.; Hora, Maninder
CS Department of Formulation Development, Chiron Corporation, Emeryville, CA, 94608-2916, USA
SO Journal of Pharmaceutical Sciences (1999), 88(9), 881-888
CODEN: JPMSAE; ISSN: 0022-3549
PB American Chemical Society
DT Journal
LA English
CC 63-5 (Pharmaceuticals)
AB Study of recombinant human tissue factor pathway inhibitor (rhTFPI) **solubility** shows (1) an inverted bell-shaped pH-**solubility** profile with a broad **solubility** min. between pH 5 and 10 such that the **solubility** min. midpoint is 2-3 pH units away from its isoelec. point; (2) a neg. temperature-**solubility** coefficient; (3) a strong dependence of **solubility** on the valence of electrolytes, with both multivalent cations and anions enhancing this effect; and (4) a significant increase of **solubility** in the presence of charged polymers. At pH 6-7, rhTFPI **solubility**-salt profiles display typical salting-in and salting-out biphasic effects. At a slightly lower pH (pH 5), a third phase in addition to the salting-in and salting-out phases was observed at low ionic strength

conditions (5 to 50 mM) where rhTFPI **solubility** increased as salt concentration decreased. The salting-out constant for rhTFPI in NaCl is 1.04 M-1 and is independent of the pH of the solution **Resolubilization** of rhTFPI ppts. revealed that "**insoly.** ppts." (seen during buffer exchanges) resulted from protein solute saturation and could be redissolved by "native" solvent conditions. On the other hand, "instability ppts." (typically seen after exposure to elevated temps. or extended storage periods) were caused by insol. protein aggregate formation and required strongly denaturing conditions to redissolve.

ST **soly** tissue factor pathway inhibitor

IT Ionic strength
Solubility
Solubilization
(**solubility** of recombinant human tissue factor pathway inhibitor)

IT Polyoxyalkylenes, properties
RL: PRP (Properties)
(**solubility** of recombinant human tissue factor pathway inhibitor)

IT 50-70-4, Sorbitol, properties 56-40-6, Glycine, properties 56-45-1, Serine, properties 56-84-8, L-Aspartic acid, properties 56-86-0, L-Glutamic acid, properties 56-87-1, L-Lysine, properties 57-13-6, Urea, properties 57-50-1, Sucrose, properties 69-65-8, Mannitol 73-32-5, L-Isoleucine, properties 77-92-9, Citric acid, properties 87-69-4, Tartaric acid, properties 110-15-6, Succinic acid, properties 110-16-7, Maleic acid, properties 127-09-3, Sodium acetate 320-77-4, Isocitric acid 994-36-5, Sodium citrate 3483-12-3, Dithiothreitol 6915-15-7, Malic acid 7447-40-7, Potassium chloride, properties 7487-88-9, Magnesium sulfate, properties 7632-05-5, Sodium phosphate 7647-14-5, Sodium chloride, properties 7757-82-6, Sodium sulfate, properties 7783-20-2, Ammonium sulfate, properties 7786-30-3, Magnesium chloride, properties 9003-01-4, Polyacrylic acid 9004-54-0, Dextran, properties 9005-64-5, Polysorbate 20 9005-65-6, Polysorbate 80 9042-14-2, Dextran sulfate 24937-47-1, Poly(L-**arginine** 24991-23-9 25212-18-4, Poly(L-**arginine** 25322-68-3, Peg 25513-46-6, Poly(L-glutamic acid) 26700-71-0, Poly(L-glutamine)
RL: PRP (Properties)
(**solubility** of recombinant human tissue factor pathway inhibitor)

IT **194554-71-7**, Tissue factor pathway inhibitor
RL: PRP (Properties); THU (Therapeutic use); BIOL (Biological study); USES (Uses)
(**solubility** of recombinant human tissue factor pathway inhibitor)

RE.CNT 24 THERE ARE 24 CITED REFERENCES AVAILABLE FOR THIS RECORD

RE

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L94 ANSWER 7 OF 9 HCAPLUS COPYRIGHT 2004 ACS on STN
AN 1998:331525 HCAPLUS
DN 129:23725
ED Entered STN: 03 Jun 1998
TI Variants of bone morphogenetic proteins that undergo refolding and active heterodimer formation
IN Wolfman, Neil M.; McCoy, John
PA Genetics Institute, Inc., USA
SO U.S., 18 pp., Cont.-in-part of U.S. 5,399,677.
CODEN: USXXAM
DT **Patent**
LA English
IC ICM C07K014-51
ICS C12N015-12
NCL 435069100
CC 2-3 (Mammalian Hormones)
FAN.CNT 2

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	US 5756308	A	19980526	US 1994-360914	19941221 <--
	US 5399677	A	19950321	US 1993-163877	19931207 <--
	US 5804416	A	19980908	US 1996-741589	19961031 <--
PRAI	US 1993-163877	A2	19931207 <--		
	US 1994-360914	A3	19941221 <--		

AB Mutant forms of bone morphogenetic proteins (BMP) are disclosed. The mutant forms of BMP can be produced bacterially and refolded to produce biol. active homodimers or heterodimers of BMP. A method of making such mutant BMPs is also disclosed. Thus, the Ser-63 residue of BMP-8 is replaced by His. Escherichia coli-produced mutant BMP-8 is successfully refolded and forms active heterodimeric complexes with other BMP proteins.

ST bone morphogenetic protein mutagenesis refolding
IT Bone morphogenetic **proteins**
RL: **BMF (Bioindustrial manufacture)**; **PEP (Physical, engineering or chemical process)**; BIOL (Biological study); **PREP (Preparation)**; **PROC (Process)**
(2; variants of bone morphogenetic proteins that undergo refolding and active heterodimer formation)
IT Bone morphogenetic **proteins**
RL: **BMF (Bioindustrial manufacture)**; **PEP (Physical, engineering or chemical process)**; BIOL (Biological study); **PREP (Preparation)**; **PROC (Process)**
(4; variants of bone morphogenetic proteins that undergo refolding and active heterodimer formation)
IT Bone morphogenetic **proteins**
RL: **BMF (Bioindustrial manufacture)**; **PEP (Physical, engineering or chemical process)**; BIOL (Biological study); **PREP (Preparation)**; **PROC (Process)**
(5; variants of bone morphogenetic proteins that undergo refolding and

active heterodimer formation)
IT Bone morphogenetic **proteins**
RL: **BMF (Bioindustrial manufacture)**; **PEP (Physical, engineering or chemical process)**; BIOL (Biological study); **PREP (Preparation)**; **PROC (Process)**
(6; variants of bone morphogenetic proteins that undergo refolding and active heterodimer formation)
IT Bone morphogenetic **proteins**
RL: **BMF (Bioindustrial manufacture)**; **PEP (Physical, engineering or chemical process)**; BIOL (Biological study); **PREP (Preparation)**; **PROC (Process)**
(7; variants of bone morphogenetic proteins that undergo refolding and active heterodimer formation)
IT Bone morphogenetic **proteins**
RL: **BMF (Bioindustrial manufacture)**; **PEP (Physical, engineering or chemical process)**; BIOL (Biological study); **PREP (Preparation)**; **PROC (Process)**
(8; variants of bone morphogenetic proteins that undergo refolding and active heterodimer formation)
IT Protein sequences
(of variants of bone morphogenetic proteins that undergo refolding and active heterodimer formation)
IT Solvents
(organic; variants of bone morphogenetic proteins that undergo refolding and active heterodimer formation)
IT Mutagenesis
(site-directed; variants of bone morphogenetic proteins that undergo refolding and active heterodimer formation)
IT **Protein folding**
(variants of bone morphogenetic proteins that undergo refolding and active heterodimer formation)
IT 146869-98-9P, Osteogenin (human B subunit reduced) 207870-75-5P 207870-76-6P 207870-77-7P 207870-79-9P 207870-80-2P 207870-81-3P 207870-82-4P
RL: BMF (Bioindustrial manufacture); PEP (Physical, engineering or chemical process); PRP (Properties); BIOL (Biological study); PREP (Preparation); PROC (Process)
(amino acid sequence; variants of bone morphogenetic proteins that undergo refolding and active heterodimer formation)
IT 207870-85-7P
RL: BPN (Biosynthetic preparation); BIOL (Biological study); PREP (Preparation)
(nucleotide sequence; variants of bone morphogenetic proteins that undergo refolding and active heterodimer formation)
IT 57-13-6, Urea, uses 64-17-5, Ethanol, uses 67-56-1, Methanol, uses 67-63-0, Isopropanol, uses 74-79-3, L-**Arginine**, uses 113-00-8, Guanidine
RL: MOA (Modifier or additive use); USES (Uses)
(variants of bone morphogenetic proteins that undergo refolding and active heterodimer formation)
RE.CNT 21 THERE ARE 21 CITED REFERENCES AVAILABLE FOR THIS RECORD
RE
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L94 ANSWER 8 OF 9 WPIX COPYRIGHT 2004 THOMSON DERWENT on STN
AN 1997-087056 [08] WPIX
DNC C1997-028244
TI Aqueous **formulation** of **tissue factor pathway inhibitor** - contains charged polymer, e.g. dextran sulphate, to facilitate **solubilisation**, **formulation purification** and **refolding** of protein.
DC A96 B04 D16
IN ARVE, B H; BILD, G S; CHEN, B; DORIN, G J; GUSTAFSON, M E; HALENBECK, R F; HORA, M S; JOHNSON, G V; JOHNSON, K; MADANI, H; PATTISON, G L; RANA, R K; TSANG, M; HALLENBECK, R F; HOBA, M S
PA (CHIR) CHIRON CORP; (SEAR) SEARLE & CO G D
CYC 72
PI

WO 9640784	A2	19961219 (199708)*	EN	86	C07K014-81
RW: AT BE CH DE DK EA ES FI FR GB GR IE IT KE LS LU MC MW NL OA PT SD SE SZ UG					
W: AL AM AT AU AZ BB BG BR BY CA CH CN CZ DE DK EE ES FI GB GE HU IL IS JP KE KG KP KR KZ LK LR LS LT LU LV MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK TJ TM TR TT UA UG US UZ VN					
AU 9664770	A	19961230 (199716)			C07K014-81
WO 9640784	A3	19970313 (199728)			C07K014-81
EP 837883	A2	19980429 (199821)	EN		C07K014-81
R: AT BE CH DE DK ES FI FR GB GR IE IT LI LU MC NL PT SE					
US 5888968	A	19990330 (199920)			A61K038-36
AU 713338	B	19991202 (200008)			C07K014-81
JP 11514334	W	19991207 (200008)		92	C07K014-81
AU 2000020611	A	20000615 (200036)#			A61K038-57
US 6319896	B1	20011120 (200174)			A61K038-16
US 6323326	B1	20011127 (200175)			A23J001-00
AU 759412	B	20030417 (200333)#			A61K038-57
CA 2450795	A1	19961219 (200414)	EN		C07K014-81
CA 2450797	A1	19961219 (200414)	EN		C07K014-81
CA 2450800	A1	19961219 (200414)	EN		A61K038-55
CA 2450804	A1	19961219 (200414)	EN		A61K038-55
CA 2450953	A1	19961219 (200414)	EN		A61K038-55
CA 2451969	A1	19961219 (200417)	EN		C07K014-81
CA 2451970	A1	19961219 (200417)	EN		C07K014-81
CA 2451973	A1	19961219 (200417)	EN		C07K014-81
JP 2004083591	A	20040318 (200420)		48	C07K014-81
CA 2223745	C	20040316 (200421)	EN		C07K014-81
AU 2003200506	A1	20030417 (200433)#			A61K038-57

ADT WO 9640784 A2 WO 1996-US9980 19960607; AU 9664770 A AU 1996-64770 19960607; WO 9640784 A3 WO 1996-US9980 19960607; EP 837883 A2 EP 1996-924269 19960607, WO 1996-US9980 19960607; US 5888968 A Cont of US 1995-477677 19950607, US 1996-734997 19961022; AU 713338 B AU 1996-64770

19960607; JP 11514334 W WO 1996-US9980 19960607, JP 1997-502126 19960607; AU 2000020611 A Div ex AU 1996-64770 19960607, AU 2000-20611 20000302; US 6319896 B1 Cont of US 1995-473668 19950607, Cont of US 1995-477677 19950607, Div ex WO 1996-US9980 19960607, Div ex US 1999-973211 19990611, US 1999-443098 19991118; US 6323326 B1 CIP of US 1995-473688 19950607, CIP of US 1995-477677 19950607, WO 1996-US9980 19960607, US 1999-973211 19990611; AU 759412 B Div ex AU 1996-64770 19960607, AU 2000-20611 20000302; CA 2450795 A1 Div ex CA 1996-2223745 19960607, CA 1996-2450795 19960607; CA 2450797 A1 Div ex CA 1996-2223745 19960607, CA 1996-2450797 19960607; CA 2450800 A1 Div ex CA 1996-2223745 19960607, CA 1996-2450800 19960607; CA 2450804 A1 Div ex CA 1996-2223745 19960607, CA 1996-2450804 19960607; CA 2450953 A1 Div ex CA 1996-2223745 19960607, CA 1996-2450953 19960607; CA 2451969 A1 Div ex CA 1996-2223745 19960607, CA 1996-2451969 19960607; CA 2451970 A1 Div ex CA 1996-2223745 19960607, CA 1996-2451970 19960607; CA 2451973 A1 Div ex CA 1996-2223745 19960607, CA 1996-2451973 19960607; JP 2004083591 A Div ex JP 1997-502126 19960607, JP 2003-326585 20030918; CA 2223745 C CA 1996-2223745 19960607, WO 1996-US9980 19960607; AU 2003200506 A1 Div ex AU 2000-20611 20000302, AU 2003-200506 20030214

FDT AU 9664770 A Based on WO 9640784; EP 837883 A2 Based on WO 9640784; AU 713338 B Previous Publ. AU 9664770, Based on WO 9640784; JP 11514334 W Based on WO 9640784; AU 2000020611 A Div ex AU 713338; US 6323326 B1 CIP of US 5923306, Based on WO 9640784; AU 759412 B Previous Publ. AU 2000020611, Div ex AU 713338; CA 2223745 C Based on WO 9640784

PRAI US 1995-477677 19950607; US 1995-473668 19950607;
US 1996-734997 19961022; AU 2000-20611 20000302;
US 1999-973211 19990611; US 1999-443098 19991118;
US 1995-473688 19950607; AU 2003-200506 20030214

REP EP 131864; EP 325691; EP 473564; EP 559632

IC ICM A23J001-00; A61K038-16; A61K038-36; A61K038-55; A61K038-57; C07K014-81

ICS A01N037-18; A61K009-08; A61K031-198; A61K038-00; A61K047-04; A61K047-10; A61K047-12; A61K047-18; A61K047-20; A61K047-22; A61K047-26; A61K047-30; A61K047-34; A61K047-36; A61P007-02; A61P043-00; C07K001-00; C07K001-02; C07K001-113; C07K001-14; C07K001-16; C07K001-18; C07K001-30; C07K014-745; **C12P021-00**

AB WO 9640784 A UPAB: 20020213

An aqueous **formulation** comprising **tissue factor pathway inhibitor** (TFPI) at a concentration of > 1 mg/ml and a charged polymer (CP) is new. The charged polymer (CP) is pref. a sulphated polysaccharide (such as heparin or dextran sulphate) or a polyphosphate, pref. immobilised on a solid support. Also new are: a method of modifying the **solubility** of a protein having a first domain which has a net positive charge and a second domain which has a net negative charge comprises adding an aqueous solution of CP to reduce inter- and intra-molecular interactions between the charged domains; and a method of **refolding** an improperly **folded** or denatured protein (e.g. TFPI) comprises adding CP to a solution of the protein prior to allowing the protein to **refold.** Further claimed is a pharmaceutically acceptable compsn. comprising > 0.2 mg/ml TFPI and a **solubilising** agent chosen from acetate ions, NaCl, citrate ions, isocitrate ions, glycine, glutamate, succinate ions, histidine, imidazole and SDS.

USE - The methods are partic. useful for **solubilising, formulating, purifying** and **refolding** proteins (especially TFPI) which have been engineered by genetic recombination and produced in bacterial, yeast or other cells in a form that has a non-native **tertiary structure.** TFPI is a coagulation inhibitor which has clot-inhibiting properties.

Dwg.0/28

FS CPI

FA AB; DCN
MC CPI: A12-V01; **B04-N03**; B04-N0300E; **D05-H13**; D05-H17

L94 ANSWER 9 OF 9 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED.
on STN
AN 93239715 EMBASE
DN 1993239715
TI Production and characterization of an analog of acidic fibroblast growth factor with enhanced stability and biological activity.
AU Arakawa T.; Horan T.P.; Narhi L.O.; Rees D.C.; Schiffer S.G.; Holst P.L.; Prestrelski S.J.; Tsai L.B.; Fox G.M.
CS Amgen Inc, Amgen Center,Thousand Oaks, CA 91320-1789, United States
SO Protein Engineering, (1993) 6/5 (541-546).
ISSN: 0269-2139 CODEN: PRENE
CY United Kingdom
DT Journal; Article
FS 022 Human Genetics
029 Clinical Biochemistry
LA English
SL English
AB We have used **recombinant** DNA methods to produce two forms of bovine acidic fibroblast growth factor (aFGF), one with alanine substituted for the cysteine at position 47 and the other with the Ala47 change plus the substitution of glycine for the naturally occurring histidine at position 93. Both forms were expressed at high levels in Escherichia coli and purified to near homogeneity by **solubilization** of the inclusion bodies containing the aFGF, ion exchange chromatography, refolding of the protein and hydrophobic interaction chromatography. Circular dichroic and infrared spectra suggested that the proteins are similar in secondary and **tertiary structures** and contain little or no α-helical conformations. Hydrophobic interaction chromatography showed that aFGF C47A/H93G is slightly more hydrophobic than the aFGF C47A form, suggesting that residue 93 is exposed to the solvent. Half-maximal activity in an in vitro bioassay system was reached at a 10- to 20-fold lower dose for the aFGF C47A/H93G form than for the aFGF C47A form, suggesting that alteration of this residue has an effect on the region responsible for receptor binding. Addition of 50 μg/ml heparin enhanced the in vitro activity of the aFGFs, reducing the half-maximal dose to approximately 100 pg/ml for both forms, comparable to that observed previously for basic FGF with or without heparin in this assay system.
CT Medical Descriptors:
*protein stability
amino acid substitution
article
circular dichroism
cow
escherichia coli
gene expression
hydrophobicity
infrared spectrometry
nonhuman
protein folding
protein purification
protein secondary structure
protein structure
Drug Descriptors:
*acidic fibroblast growth factor
alanine
cysteine

glycine
heparin
histidine
protein derivative
recombinant dna

RN (acidic fibroblast growth factor) 106096-92-8; (alanine) 56-41-7, 6898-94-8; (cysteine) 4371-52-2, 52-89-1, 52-90-4; (glycine) 56-40-6, 6000-43-7, 6000-44-8; (heparin) 37187-54-5, 8057-48-5, 8065-01-8, 9005-48-5; (histidine) 645-35-2, 7006-35-1, 71-00-1

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